

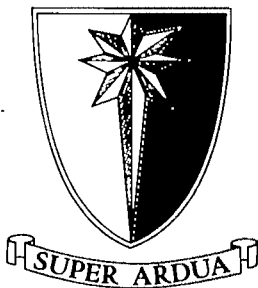
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Edited by Frank E. Hytten

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Measurements should be expressed in SI units with the exception of haemoglobin (g/dl) and blood pressure (mmHg). If the original observations were recorded in other units this should be stated, together with the appropriate conversion factors. Standard abbreviations, without punctuation, are used. *Units, Symbols and Abbreviations* (1977) published by

the Royal Society of Medicine provides a useful guide. Abbreviations are used sparingly and only if a lengthy name or expression is repeated frequently throughout the manuscript. Words must appear in full on first appearance in both summary and text, followed by the abbreviation in parenthesis.

Drugs are described by their official names but trade names should be indicated in brackets the first time a drug is quoted in the main text.

Statistics

Authors should describe clearly the design of their investigation and the source of the data. All statistical methods should be specified so that it is clear which was used where. Papers omitting this information may be returned without review. The basic principle to follow is to supply sufficient information about the design and methods of analysis to allow the research to be repeated by someone else.

Some points of detail to note are:

- (1) It is not acceptable to present the results of a statistical analysis solely by a *P* value—actual results such as means or proportions should always be given, along with the test statistic.
- (2) The use of confidence intervals is encouraged.
- (3) The symbol $\pm$ should not be used when presenting SE or SD. Presentation such as 'the mean was 14.2 (SD 3.2) mg' is preferred.
- (4) When paired comparisons are made it is desirable to indicate the variability of within-pair differences.
- (5) Unusual methods of analysis should be described and/or referenced.

Authors are referred to the general statistical guidelines of Altman *et al.* (1983) for advice about the presentation and interpretation of the statistical aspects of their research.

Altman, D. G., Gore, S. M., Gardner, M. J. & Pocock, S. J. (1983) Statistical guidelines for contributors to medical journals. *Br Med J* 286, 1489–1493.

Human experiments

Manuscripts should contain a statement that the work was approved by the ethics committee of the institution in which it was done and that subjects gave informed consent to the work. The editors reserve the right to reject papers if they consider the ethical justification to be questionable.

References

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References to 'personal communications' and 'unpublished work' may be quoted in the text but should not be included in the references. Examples of the style to be used are given below.

Trudinger, B. J., Lewis, P. J. & Pettit, B. (1979) Fetal breathing in intrauterine growth retardation. *Br J Obstet Gynaecol* 86, 432–436.

Hyttén, F. E. & Leitch, I. (1971) *The Physiology of Human Pregnancy* 2nd edn, Blackwell Scientific Publications, Oxford, p. 26.

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COMMENTARY

Fetal rights

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A 36-year-old woman, referred to as F, became pregnant early in 1987. She was under the care of a consultant psychiatrist, having a history of severe mental disturbance and periodic drug abuse. Her circumstances were well known to local social services officers; her 10-year-old son had been taken into care some years before, and her access to him had been terminated as part of long-term foster arrangements. F continued her appointments until January 1988 when, just before the expected date of the birth of the fetus, she disappeared. The local authority applied for the fetus to be made a ward of court to protect its welfare. This is the first case in which this step is known to have been initiated in the UK.

The effect of a wardship order is to terminate all formal parental rights, duties and powers over the child and vest them in the court. In F's case the local authority wanted the court to order the tipstaff to find and detain her, require her to surrender her passport and not attempt to take her child away after its birth, and finally, to order her to attend a hospital nominated by the social services. On birth, the authority would require F to comply with further instructions as to the child's care and control. According to one judge, Lord Justice Staughton, these were orders 'which seek directly to control the life of both mother and child'. A High Court judge, Justice Hollins, refused the authority's request. It appealed to a panel of three more senior judges in the Court of Appeal; Lords Justice May, Staughton and Balcombe. They dismissed the social workers' case.

This is at least the fourth recent example in which a social work department is known to have contemplated the use of wardship in seeking protection of a fetus. In one case, the authority anxiously discussed the possibility of using wardship to force a mentally competent woman to submit to a caesarean section. Although the Court of Appeal has said clearly that courts cannot extend the use of the wardship jurisdiction to

protect the interests and welfare of a fetus, Lord Justice May indicated that this was certainly a case in which it would have been justified. F's case bears the hallmark of an emerging parallel with legal developments in the USA. There, court-ordered obstetrical interventions have sanctioned non-consensual, non-therapeutic caesarean sections, compulsory hospitalization and intrauterine transfusions. The spectres of judicial support for prenatal screening, fetal surgery and restrictions on the diet, work activity, and athletic and sexual recreations of pregnant women are feared to be waiting in the delivery room. Some (Leiberman *et al.* 1979) will warmly welcome the arrival of these judicial spirits as welfare guardians, but once they are in house, stalking and exorcizing the ghosts of legal supervision will be troublesome and harrowing.

All the judges who heard this case agreed that, until birth, the fetus has no civil legal personality. Whatever its maturing moral character and physiological status, the fetus enjoys only the indirect protection afforded by the law on abortion. English law draws a fundamental distinction between a newborn infant and a fetus. A claim to inherit property, or in respect of injuries inflicted before birth 'crystallizes upon the birth, at which date, but not before, the child attains the status of a legal persona, and thereupon can then exercise that legal right' (Lord Justice May). The wardship jurisdiction of the court did not extend to the fetus; 'since an unborn child has, *ex hypothesi*, no existence independent of its mother, the only purpose of extending the jurisdiction to include a fetus is to enable the mother's actions to be controlled' (Lord Justice Balcombe). Respecting the autonomy of the woman outweighed any claims which might have been advanced on behalf of the fetus.

The court was unimpressed with the argument that orders of the kind sought could be limited to viable fetuses, so as not to conflict with the interests of women seeking an early abortion. Lord

Justice Balcombe said that if the wardship jurisdiction extended to the fetus, there was no logic, although there might be sound pragmatism, in limiting it only to the viable fetus. The abortion provisions could not be confined in that way, and neither could the wardship jurisdiction. He was similarly dismissive of an argument based on Article 2(1) of the European Convention for the Protection of Human Rights and Fundamental Freedoms. This provides that 'Everyone's right to life shall be protected by law'. According to Lord Justice Balcombe, extending this for the benefit of the fetus would mean that a pregnant woman's 'right to life' would be subject to an implied limitation in favour of the fetus. In an extreme case, such as a woman needing an abortion in order to avert a serious threat to her life, this could entail sacrificing the mother's life to preserve the right of the fetus. The court refused to accept this conclusion.

Clearly, all the judges were concerned about the consequences of extending the jurisdiction. Lord Justice Balcombe was alarmed that the court might be asked to exercise its inherent jurisdiction to control and direct a woman's health regimen (no smoking, no alcohol), social life ('any activity which might be hazardous to the child'), and possibly her obstetrical preference. The judges agreed that if the wardship jurisdiction needed extension to protect the fetus, then that was a parliamentary and not judicial responsibility. The court had no guidance as how such a responsibility should be exercised and discharged.

Some lawyers have regarded the use of the wardship jurisdiction to protect the fetus as 'a logical and natural development' of the powers of the courts. As a form of defensive social work practice these recent moves can be understood as attempts to pre-empt criticism that local authorities have not anticipated danger to a new client group, unborn fetuses and newborn infants. They reflect and are a representative part of a more general mood which has seen the emergence of the fetus, figuratively and literally, from the shadows of moral concern. They are part of a broader movement to accord rights to the unborn, which is paralleled in the concern expressed about human embryo research. To date, the vantage of British judges has resembled that of the Wimbledon spectator; clearly having their own private view as to which shots are out of court, but deferring to the legislative calls. But as some of the cases get closer to

lines set by social priorities of different times, courts will get drawn more closely into reproductive conflicts. There are, however, at least two cardinal reasons why I think that temptation was correctly resisted in *Re F*; the 'slippery slope' argument, and the 'therapeutic conflict' consideration.

These may undoubtedly be regarded as difficult and troubling cases. But they must be recognized to be at the top of a tortuous and slippery slope. The 'horrible result' which lies at the bottom is the translation of pregnant women into nothing more than 'fetal containers' (Annas 1986), seeing them as no more than a resource to be used to the ends of another. English law has always been remarkably reticent about the imposition of a positive duty to come to another's rescue. It would be inappropriate to accord more respect to the integrity of a human corpse, which might yield organs or tissue for a life-saving transplant, than to the autonomy of a pregnant woman; yet English law presently allows the surviving relatives of a deceased person effectively to frustrate even the express wishes announced by the deceased before death (Human Tissue Act 1961, as amended).

Alternatively, the 'arbitrary result', once on the slippery slope, concerns the appropriate fetal age; what sort of threats the fetus must be exposed to; what likelihood of harm is necessary; and so on, before the court would entertain wardship. And if enforced medical regimens were countenanced, what potentially welfare threatening would not be regulated? (Williams (1985) distinguishes these two types of argument.)

Secondly, lawyers should be hesitant to rush into the delivery room. The relationship between pregnant women and obstetricians is not straightforward; creating an adversarial ground between the woman and her fetus will not clarify it. Physicians often disagree about the appropriateness of obstetrical intervention, and the intrusion of professional social workers and lawyers into the doctor-patient relationship would be a doubtful way of resolving any gathering conflict. In fact, most women when faced with the risk of death or serious injury to their fetus, do, even if begrudgingly, submit to their obstetrician's judgement. While English law is quick to accept medical uncertainty as a justification for physician error, we seem in danger of being less ready to recognize the implications of

this for patient self-determination; this point is forcefully made by Kolder *et al.* (1987).

If there is a course to be set between the Scylla of treating women as fetal containers and the Charybdis of regarding their fetuses as uterine cargo (Meeker 1987), the price which we must be prepared to pay for protecting the integrity and autonomy of all competent adults is the rare, occasional risk of death or serious injury to an unborn fetus. It cannot be that it is professionals alone who are entitled to make the occasional mistake.

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Genetic control of susceptibility to eclampsia and miscarriage

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Summary. An analysis has been made of 48 pedigrees selected (ascertained) through an affected mother in the first generation. These pedigrees mainly involve cases of eclampsia which occurred before its recent decline in incidence. The data confirm the genetic determination of susceptibility indicated by published data on eclampsia/pre-eclampsia. There is a suggestion that the fetal genotype can contribute to susceptibility to eclampsia in its mother, in contrast to previous findings that susceptibility to pre-eclampsia is controlled solely by the maternal genotype. An association between eclampsia and miscarriage is shown in the data. We argue that this suggests that the primary mode of action of the gene(s) involved is to affect the interaction between uterine and placental tissue.

Eclampsia/pre-eclampsia is a disease which has been known since antiquity (Chesley 1978; Macgillivray 1983). While its physiological and biochemical cause remains to be discovered, there is considerable evidence which suggests that it is largely genetic in origin. Many reports of individual pedigrees with a number of cases of eclampsia have appeared during the last century (summarized by Chesley *et al.* 1968). However,

the long human generation time combined with the confinement of the phenotype to pregnancy render a study of its genetic control rather difficult and only one systematic attempt has been made. Chesley *et al.* (1961, 1968) studied the prevalence of pre-eclampsia and eclampsia in the female relatives of 187 eclamptic women. The frequency of both pre-eclampsia and eclampsia in the sisters and daughters of the index cases was higher than that in the general population. Chesley's data and data from Scotland (Adams & Finlayson 1961; Cooper & Liston 1979) show that susceptibility to pre-eclampsia is highly heritable and is possibly under simple genetic control.

Since eclampsia/pre-eclampsia occurs only in pregnancy it is not clear whether the genotype of the woman or her fetus determines susceptibility. In principle, susceptibility could be controlled by the maternal genotype alone, or the fetal genotype alone, or by a genotype-by-genotype interaction between the mother and her fetus. Data which compare the frequency of pre-eclampsia in mothers versus mothers-in-law of pre-eclamptic index cases (Sutherland *et al.* 1981) and pre-eclampsia in daughters versus daughters-in-law of eclamptic index cases

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(Chesley & Cooper 1986) show that the maternal genotype alone is mainly responsible for susceptibility to pre-eclampsia. No effect of the fetal genotype, or of a fetal $\times$ maternal genotype interaction can be detected in these data. But data showing increased HLA antigen sharing between affected women and their spouses (Jenkins *et al.* 1978) and increased HLA homozygosity in another series of affected women and their spouses (Redman *et al.* 1978) are only interpretable in terms of a fetal genotypic influence.

Most of the data upon which these conclusions are based were collected after 1950. From about this time the introduction of present-day antenatal practices greatly reduced the frequency of eclampsia. Many cases which might have progressed to eclampsia did not do so, and are therefore recorded as pre-eclampsia. Here we report eclampsia pedigree data from Georgia, USA, collected before this decline in frequency. The data confirm the high heritability of eclampsia and give an important clue to the mode of action of the gene(s) for susceptibility.

Patients and methods

The primary source of information was the files originally compiled by Dr Bryans on women who had had eclampsia between 1919 and 1946 (Bryans & Torpin 1949). All information contained therein was verified, supplemented, or modified by marriage and birth registration in the state registry of vital statistics in either Augusta or Atlanta, Georgia, USA, by three of us (J.A.H., L.C.C., C.I.B.). Children of the index cases were discovered by birth registration. These names were then cross-referenced with marriage registries. Once the married name was found both it and the maiden name were used to find offspring from birth registration. A similar procedure was used to find their marriage and their offspring, hence giving names of children, grandchildren, great grandchildren, and great great grandchildren of the index cases. Using the names found from the registries, the medical record department of hospitals in the Augusta region (Medical College of Georgia, University Hospital, St Joseph's and Humana) were searched for corresponding charts. When the charts were located, information concerning pregnancies was recorded to determine whether the patient had pre-eclampsia or eclampsia.

In general, the information recorded in the

charts enabled us to determine whether or not the patient had elevated blood pressure, proteinuria, oedema, or seizure. The diagnoses of eclampsia are likely to be reliable, since the seizures cannot easily be confused with other conditions. The distinction between 'true' pre-eclampsia and other hypertensive disorders in pregnancy is much more suspect. Some at least of those recorded as having pre-eclampsia may in reality have had essential hypertension or renal disease. Since the diagnosis of pre-eclampsia in first pregnancies on the basis of these characteristics is more likely to be correct in first than in later pregnancies (Fisher *et al.* 1981), only those recorded as having pre-eclampsia in first pregnancy have been treated as such in our analysis.

All 50 eclamptic index cases survived their seizure. They led to 48 pedigrees. Of these, 46 were singly selected (ascertained) through an eclamptic mother in generation I, one was doubly selected through a mother in generation I and her daughter, and one through a mother in generation I and her daughter-in-law. There were 22 black pedigrees (23 index cases), in 20 of which the index case had a daughter who reproduced at least once, and 26 white pedigrees (27 index cases), with 24 having a daughter who reproduced at least once. One index case had eclampsia in her first and second pregnancies; the remainder had only one eclamptic pregnancy.

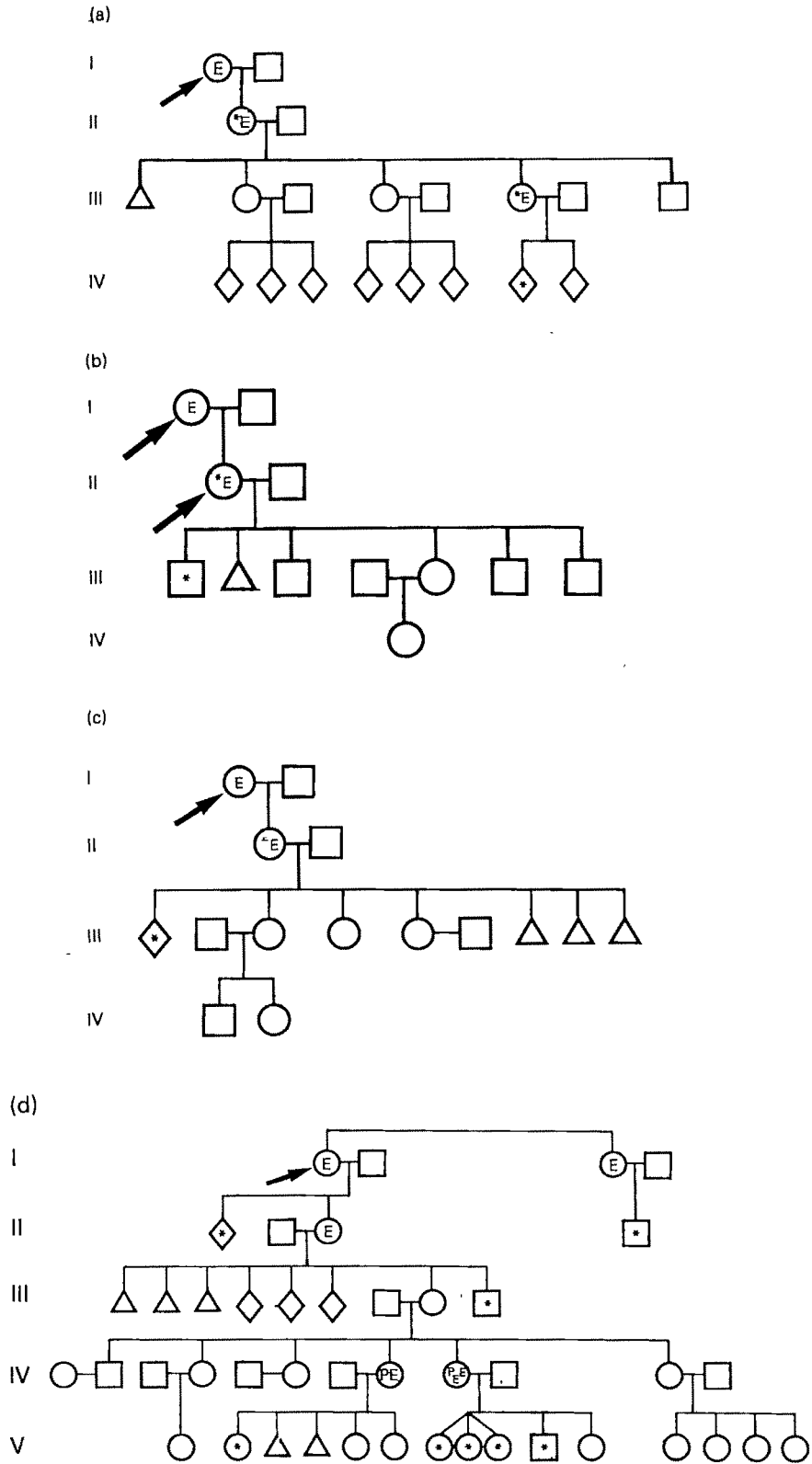
The family data given are from families for which there was complete information, i.e. information was obtained on the outcome of all the pregnancies of all daughters in the family. All pedigrees have been recorded on microfilm and are available from D.W.C

Results

Some illustrative pedigrees are shown in Fig. 1. Eighteen of the 48 pedigrees had at least one example of mother and daughter with eclampsia. Thirty pedigrees extended over four or more generations, so that one at least of the members of the third generation was tested for her susceptibility to eclampsia. Three cases of eclampsia occurred in the third generation.

Daughters of eclamptic index cases

Most of the data are for this class of offspring. Table 1 shows the number of normal, pre-



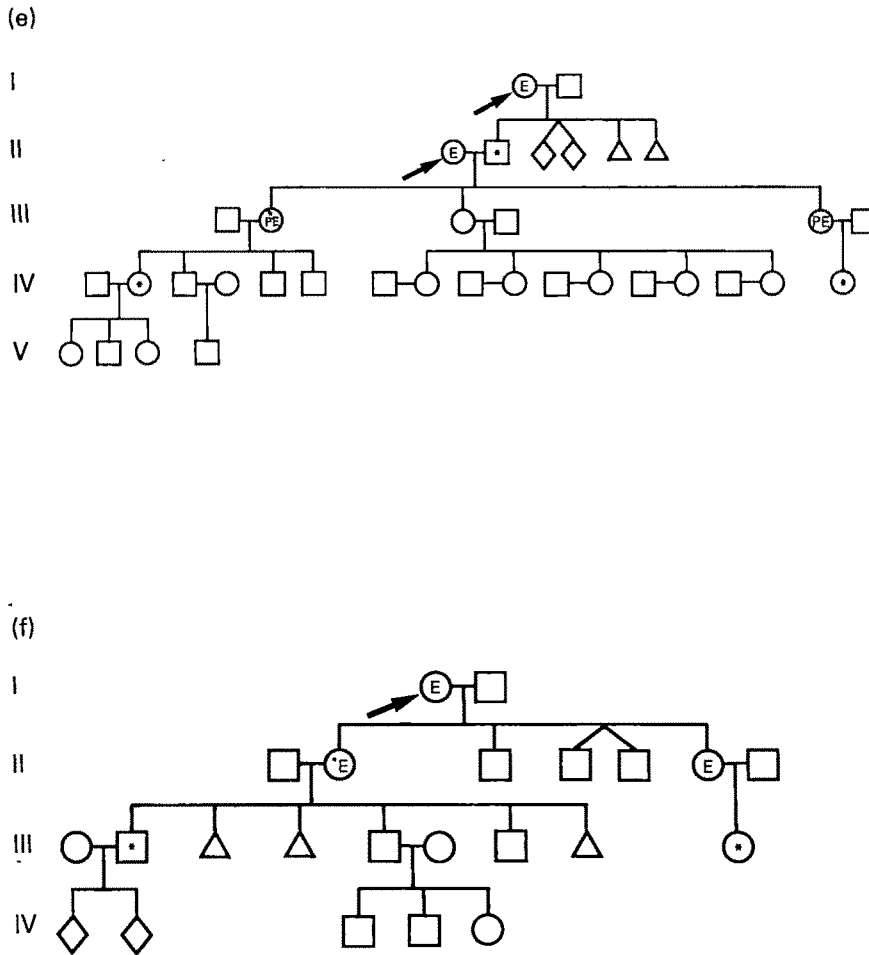


Fig. 1. Examples of eclampsia pedigrees in black and white Americans. □, male; ○, female; ◇, sex unknown; △, miscarriage; E, eclampsia; PE, pre-eclampsia; *, child born of eclamptic or pre-eclamptic pregnancy, →, index case(s) in each pedigree. (a) Black pedigree showing eclampsia in three successive generations, the only such example of this in the data set. Note also miscarriage followed by eclampsia in generation II/III. (b) White pedigree showing eclampsia followed by miscarriage. There are two index cases in this pedigree. (c) White pedigree also showing occurrence of miscarriage and eclampsia. (d) Black pedigree showing occurrence of eclampsia in sisters. Eclampsia occurs in generations I, II and IV. The women in generation IV who had both pre-eclampsia and eclampsia suffered the former when pregnant with triplets and the latter with the singleton in her second pregnancy. (e) Black pedigree showing double ascertainment and eclampsia in a daughter-in-law. (f) White pedigree with eclampsia in two daughters of the index case.

Table 1. Distribution of eclamptic, pre-eclamptic, and normal daughters in offspring of eclamptic women

Eclamptic mothers	No. of eclamptic mothers*	Daughters			Total
		Normal	Pre-eclamptic only†	Eclamptic	
Black index	21	18	5	8	31
Black non-index	8	7	4	1	12
White index	24	25	5	9‡	39
White non-index	2	2	0	0	2
Totals	55	52	14	18	84
Percentages		62	17	21	100

* Five of the original index cases did not have any daughters with full-term pregnancies

† In first pregnancy (see comment in Materials and methods).

‡ Including one eclamptic daughter who was herself an index case.

Index cases are the eclamptic women through whom the pedigree was originally selected. The non-index cases are the other eclamptic women in these pedigrees, i.e. for the most part the daughters and granddaughters of the index cases

eclamptic and eclamptic daughters of eclamptic women. The data for black and white women are very similar; when combined they show that 20% (17/84) of daughters suffered eclampsia (the eclamptic daughter who was herself an index case is not counted), and a further 16% (14/84) were recorded as having pre-eclampsia in their first pregnancy. The rate of eclampsia in the daughters of index cases is non-significantly higher than in the daughters of non-index cases, 17/70 (24%) versus 1/14 (7%) ($\chi^2 = 2.03$, $P > 0.05$). All but one of the cases of eclampsia occurred before 1960 (Table 2). Most cases occurred in first pregnancy, 56/67 (84%) (Table 3). The numbers of completed pregnancies for both black and white eclamptic daughters of the index cases are higher than for their normal and pre-eclamptic sisters (Table 4). These data, of course, refer to women who have had at least one pregnancy, and were extracted only to show the extent to which the daughters of eclamptic women had been tested for their susceptibility to eclampsia. (They are not suitable for comparing

the number of children eclamptic and non-eclamptic women have in a lifetime.)

Table 5 shows that the proportion of eclamptic daughters of eclamptic women is much higher if the daughter was herself born of the mother's eclamptic pregnancy. Again the data for black and white women are similar; the combined figures are 10/27 (37%) versus 8/57 (14%) ($\chi^2 = 41.4$, $P < 0.001$). The proportion of pre-eclamptic daughters of eclamptic women is also higher if the daughter was born of the eclamptic pregnancy, 8/27 (30%) versus 6/57 (11%).

As illustrated in Fig. 1, a high proportion of recorded miscarriages was noted in the pregnancies of the eclamptic index cases. A comparison of the incidence of miscarriage in eclamptic daughters and their normal and pre-eclamptic sisters was therefore made (Table 6). Since it is necessary for a woman to have at least two pregnancies to be tested for susceptibility to both miscarriage and eclampsia, the percentage of such women who had had a miscarriage was calculated for the eclamptic index and eclamptic non-index groups and their daughters. Clearly,

Table 2. Decades in which women had eclampsia

Group	1919-1930	1930-1940	1940-1950	1950-1960	1960-1980	Total
Black index cases	12	11	1	0	0	24
White index cases	12	13	1	0	0	26
Total index cases	24	24	3	0	0	50
Black progeny	0	1	4	1	1	7
White progeny	0	0	2	6	0	8
Total progeny	0	1	6	7	1	17

Table 3. Relation between parity and occurrence of eclampsia in black (B) and white (W) women

Number of pregnancies	Pregnancy in which eclampsia occurred										Totals	
	1st		2nd		3rd		4th		5th			
	B	W	B	W	B	W	B	W	B	W	B	W
1	10	9									10	9
2	2	4	0	1							2	5
3	4	4	1	0	0	0					5	4
4	3	8	0	1	1	0	1	0			5	9
≥5	5	7	2	0	1	0	1	1	1	0	10	8
Totals	24	32	3	2	2	0	2	1	1	0	32	35
Totals for black and white women	56		5		2		3		1		67	
Percentages	84		7		3		4		1		99	

on this basis, the percentage of eclamptic women who have a miscarriage is much higher than for their non-eclamptic daughters or sisters. Specifically, the figures for all eclamptic women are 21/49 (43%) versus 2/45 (4%) for all non-eclamptic women; the difference is significant, $\chi^2_1 = 18.7$, $P < 0.01$. Similarly, amongst the eclamptic daughters of eclamptic index cases 6/11 (55%) had at least one miscarriage whereas only 1/19 (5%) of their non-eclamptic sisters did (Table 6). The difference is again significant ($P = 0.004$).

Other female relatives

No attempt was made to obtain complete information on sisters or daughters-in-law of index cases, but five of the 50 index cases had sisters who also had eclampsia. Information was available for 12 daughters-in-law of eclamptic

women: two had eclampsia, and one of these was herself an index case.

Discussion

The genetic analysis of eclampsia can present several problems. The main one in our investigation concerns the selection (ascertainment) of pedigrees. The data all ultimately derive from hospital records, and so information on daughters whose pregnancies were normal is less likely to have been collected than on daughters with complicated pregnancies. It was possible to follow up only 48/456 of the original index cases. Of these 456, 15% died as a result of their eclampsia and follow-up was not possible. Thus 48/386 (12.4%) of the survivors were followed up. The true proportion of eclamptic daughters from eclamptic index cases could be as low as 2.6% ($21\% \times 0.124$) if all of the 338 survivors who

Table 4. Number of completed pregnancies per women in eclamptic cases and their daughters

Group	Mean	Range	Variance of mean	Number of cases
Black eclamptic index cases	3.61	1-9	0.45	23
White eclamptic index cases	3.22	1-8	0.37	27
Black eclamptic non-index cases	3.0	1-7	0.76	9
White eclamptic non-index cases	3.13	1-5	0.52	8
Black pre-eclamptic daughters*	2.22	1-5	0.55	9
White pre-eclamptic daughters*	2.00	1-3	0.71	5
Black normal daughters*	2.72	1-7	0.32	25
White normal daughters*	2.48	1-6	0.28	27

* Of both eclamptic index and non-index cases

Table 5. Eclampsia and pre-eclampsia in the daughters of eclamptic women classified according to whether or not they were born of their mother's eclamptic pregnancy

Group	Daughters of:	Normal	Pre-eclamptic only	Eclamptic	Total
Born of eclamptic pregnancy	Black index	2	3	3	8
	Black non-index	2	3	1	6
	White index	5	2	6	13
	White non-index	0	0	0	0
	Total	9	8	10	27
	Percentages	33.3	29.6	37.0	
Not born of eclamptic pregnancy	Black index	16	2	5	23
	Black non-index	5	1	0	6
	White index	20	3	3	26
	White non-index	2	0	0	2
	Total	43	6	8	57
	Percentages	75.4	10.5	14.0	

escaped follow-up had daughters with normal pregnancies.

Failure to find records of daughters' pregnancies could be due to various reasons, of which the most probable are: (i) failure of the daughters to reproduce, (ii) production of all male families by the index case, (iii) migration of the mother or daughter from the Augusta area, (iv) inadequate information in the records to allow the daughter to be traced and (v) reproduction in the Augusta area but not in one of the four hospitals from which information was obtained. Of the five pos-

sibilities, only the last is likely to contribute to selection bias, since these daughters are presumably more likely to have had normal pregnancies. Thus the unbiased figure for the proportion of eclamptic daughters produced by all 386 surviving eclamptic mothers is probably considerably greater than 2.6% but somewhat less than 21%.

The rates of eclampsia in obstetric services in various parts of Europe, the USA and New Zealand have been in the range of 0.25% to 0.073% up until about 1960, with black and white

Table 6. Recorded miscarriages of eclamptic index cases and their eclamptic and non-eclamptic daughters

Group	Total women	Total with two or more pregnancies* (A)	Number of miscarriages			Total with miscarriages (B)	B/A (%)
			1	2	3		
Black eclamptic index	23	18	1	4	2	7	39
White eclamptic index	27	20	7	1	0	8	40
Total eclamptic index	50	38	8	5	2	15	40
Black eclamptic non-index	9	5	1	0	1	2	40
White eclamptic non-index	8	6	2	0	2	4	67
Total eclamptic non-index	17	11	3	0	3	6	55
Black non-eclamptic daughters of eclamptic index cases	16	10	1	0	0	1	10
White non-eclamptic daughters of eclamptic index cases	18	9	0	0	0	0	0
Black non-eclamptic daughters of eclamptic non-index cases	18	13	0	1	0	1	8
White non-eclamptic daughters of eclamptic non-index cases	14	13	0	0	0	0	0
Total non-eclamptic daughters	66	45	1	1	0	2	4

* Of which at least one was full term.

Table 7. Comparison of the prevalence of eclamptic, pre-eclamptic, and normal daughters of eclamptic women in data of Chesley* and this paper

	Normal	Pre-eclamptic only	Eclamptic	Total
Chesley data* (%)	183 (73.8)	58 (23.4)	7 (2.8)	248
This paper (%)	52 (61.9)	14 (16.7)	18 (21.4)	84

* Chesley & Cooper (1986)

American women having similar values (Chesley 1978). Direct estimates of the prevalence of women who have had at least one eclamptic pregnancy do not exist, but they cannot be more than an order of magnitude higher than the general prevalence, i.e. the prevalence per pregnancy. It is clear, therefore, that the prevalence of eclamptic daughters of our index cases must be far higher than the population values.

Table 7 compares our data with those of Chesley & Cooper (1986). Their data were collected from an urban area (New Jersey) on average about a decade later than ours. There is a 10-fold difference in the frequency of eclampsia in daughters of the index cases. The combined rate of eclampsia and pre-eclampsia is higher in our data—36.6% versus 25.4%. Chesley was able to follow up almost all (96%) of the families of his original index case eclamptic patients and obtained information on the pregnancies of all daughters of these families. Thus his data probably give unbiased estimates for the population he studied and the differences between his data and ours are mainly due to selection bias. In particular, the higher figure for the combined prevalence of eclampsia plus pre-eclampsia in our data could be explained as the result of a selection bias towards the inclusion of families with a higher prevalence of eclampsia. The higher figure for eclampsia alone is presumably partly explained by such selection bias and partly by the earlier period of collection of our data.

Both sets of data agree in showing that the condition eclampsia/pre-eclampsia is highly heritable. Our inability to specify the extent of the selection bias makes it inappropriate to use our data to test particular models of genetic control. Analyses of Chesley's data (Cooper & Liston 1979; Chesley & Cooper 1986) and of Scottish data (Sutherland *et al.* 1981) show close agreement with the hypothesis that susceptibility to eclampsia/pre-eclampsia is due to a single recessive gene with a frequency of about

0.25 in white populations. These data implicate only the maternal genotype.

In our data, however, there is some indication that susceptibility to eclampsia is influenced by the fetal genotype. If the fetal genotype does contribute towards susceptibility, women born of an eclamptic pregnancy are more likely to have the predisposing genotype than their sisters born of normal pregnancies. Thus the raised prevalence of eclampsia in daughters born of an eclamptic as opposed to a non-eclamptic pregnancy (Table 5) could be taken to indicate an influence of the fetal genotype on susceptibility to eclampsia, although the data do not firmly establish this conclusion. A demographic explanation can account for at least part of the observed difference. Most cases of eclampsia occurred in first pregnancy (Table 3) and so women born of such a pregnancy are mostly older than their siblings born of the non-eclamptic pregnancy. These women are accordingly more likely to have reproduced at a time when eclampsia had a higher frequency than was the case with their non-eclamptic siblings.

Because HLA data on antigen sharing and homozygosity in affected women and their partners (Jenkins *et al.* 1978; Redman *et al.* 1978) also suggest a fetal genotypic influence, more definitive data must be sought in order to assess the relative contributions of maternal and fetal genotypes to susceptibility. The series reported by Jenkins *et al.* (1978) and Redman *et al.* (1978) contained many women with severe pre-eclampsia; it is reasonable to suppose that these women might have proceeded to eclampsia had they not received the benefit of modern treatment. Since pre-eclampsia is much more common than eclampsia, we suggest that most cases of pre-eclampsia are determined by the maternal genotype, but that if the condition is to proceed to eclampsia, or what would be with modern treatment severe pre-eclampsia, an added contribution from the fetal genotype is necessary. The genes acting in the fetus are presumably the HLA genes themselves or are linked to HLA.

Selection bias is unlikely to be the explanation for the differences in occurrence of miscarriage between the eclamptic women and their normal or pre-eclamptic daughters. The data (Table 6) strongly suggest that susceptibility to eclampsia is highly correlated with susceptibility to miscarriage.

Young (1927) hypothesized a 'toxemia sequence' and 'abortion taint' in which pregnancies, before and after eclampsia, were complicated by miscarriage, premature deliveries, placental abruption and recurrent hypertension. Chesley (1978) compared his own data on the frequency of miscarriage in pregnancies following eclampsia with those of Bryans and Torpin (1949). He concluded that neither series showed convincing evidence in favour of Young's hypothesis as far as miscarriage was concerned, although the absence of control groups left the question open. In our data the information on miscarriage in non-eclamptic siblings provides a fairly stringent control group, and shows that, despite the small numbers, the hypothesis must, after all, be taken seriously. Another hint of a possible connection between eclampsia and miscarriage is found in the increased HLA antigen sharing which has been demonstrated in couples with recurrent miscarriage (reviewed in Thomas *et al.* 1985).

If true, the increased rate of miscarriage would represent the first detectable manifestation of the eclamptic genotype in pregnancy, since most of the miscarriages presumably occurred in the first trimester. It would suggest that the most likely mode of action of the predisposing genes involves the interaction between the uterine and trophoblast tissue. A similar suggestion has been made on anatomical grounds by others. Robertson *et al.* (1985) summarized evidence for their view that pre-eclampsia is associated with abnormal or retarded development of the spiral arteries; they say that in pre-eclamptic pregnancies 'the second wave of endovascular trophoblast migration does not occur in second trimester, leaving the myometrial segment of the spiral (uteroplacental) arteries as normal high resistance vessels ... responsive to maternal vasomotor changes'. Acute atherosclerosis, characteristic of pre-eclampsia, then results. Hustin *et al.* (1983) also noted acute atherosclerosis in the decidual segments of uteroplacental arteries in placental bed biopsies taken during caesarean section of pre-eclamptic women. They argued from data on the deposition of immunoglobulin

and complement components on the walls of uteroplacental arteries that an immunological mechanism might be involved in the pathogenesis of acute atherosclerosis. Our data suggest that these lines of investigation should be pursued further. It would also be worthwhile to examine genetic variation restricted to the decidua and other uterine components, to see if any of it is correlated with susceptibility to eclampsia/pre-eclampsia.

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Short-term reproductive prognosis when no cause can be found for recurrent miscarriage

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Summary. The short-term reproductive prognosis of recurrent miscarriage for which no cause was found has been evaluated in 95 couples investigated between 1980 and 1986 at the First Obstetric and Gynaecological Clinic of the University of Milan. The actuarial overall 3-year livebirth delivery rate was 64%, increasing constantly with time. The reproductive success rate decreased with the number of previous miscarriages from 80% in women with two, to 60% with three and 46% with four or more miscarriages. No effect of age and socio-economic status emerged. There was a positive association between the number of previous miscarriages and the risk of miscarriage in the next pregnancy. Compared with women with two miscarriages the relative risk of another miscarriage was 2.3 for those with three previous miscarriages and 5.0 for those with four or more (χ^2 for trend adjusted for age = 5.2, $P = 0.02$).

Among women with a history of recurrent miscarriage, no cause can be found in about 40% so that no specifically helpful therapy can be recommended. However, these women would be helped by knowing what chance they have of a successful outcome in a future pregnancy. Studies of recurrent miscarriage show marked differences in the probability of the next pregnancy producing a viable infant. Malpas (1938) and Eastman (1946) suggested that the chance of a livebirth was about 30%. More recently rates of 50-70% have been reported (Lamb 1972; Tho

et al. 1979; Harger *et al.* 1983; Stray-Pedersen & Stray-Pedersen 1984). These differences may be explained by the small numbers of women studied and deficiencies in long-term follow-up. Further, the effect of maternal age and previous abortions on the prognosis for the next pregnancy has not been properly investigated.

We have studied a consecutive series of women in whom no cause could be found for their recurrent miscarriages. Our patients came from a population that differs in parity and general lifestyle habits from those studied by others in Northern Europe and North America.

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Patients and methods

Between January 1980 and January 1986, 158 women who had had two or more miscarriages before 28 completed weeks gestation without previous term pregnancies were studied at the First Obstetric and Gynaecological Clinic of the University of Milan.

Within 6 months after their last miscarriage all the women had an assessment of their general

Table 1. Distribution of 158 couples with recurrent miscarriages according to their aetiological factors, Milan, Italy, 1980–1986

Aetiological factor	Patients (<i>n</i> = 158)	
	No.	Per cent
Mullerian		
Uterine	28	18
Cervical	19	12
Endocrine	8	5
Genetic	8	5
Unknown	95	60

Table 2. Main characteristics of 95 women with recurrent miscarriage with no recognized cause, Milan, Italy, 1980–1986

Characteristic	No.	Per cent
Age (years)		
<24	11	12
25–29	20	21
30–34	41	43
35–39	18	19
≥40	5	5
Previous miscarriages		
2	43	45
3	29	31
≥4	23	24
Social class		
IV or V (lowest)	20	21
III	47	49
I or II (highest)	12	13
Undefined	16	17

health, a hysterosalpingogram, a luteal phase endometrial biopsy, a glucose tolerance test, a hormonal profile (three progesterone and prolactin assays in the luteal phase plus one T₃, T₄, TSH, FTI assay), and both the woman and her partner had their karyotype determined from peripheral leucocytes.

Table 1 lists the frequency of the different detected causes for the recurrent miscarriages in the 158 couples. No abnormalities were detected in 95 (60%) of the couples and the outcome of further pregnancies in this group is the subject of this study. The general characteristics and reproductive history at diagnosis of these patients are shown in Table 2.

During a subsequent pregnancy each woman was advised to rest as much as possible, to avoid heavy work and travelling, and she was offered a weekly medical consultation. Beta-mimetic

drugs were prescribed on clinical criteria after 15 weeks gestation in 22 (23%) and in three women cerclage was performed between 12 and 20 weeks gestation. There was no perinatal mortality and no fetal abnormality in the pregnancies studied. The present analysis is based on data obtained up to November 1986.

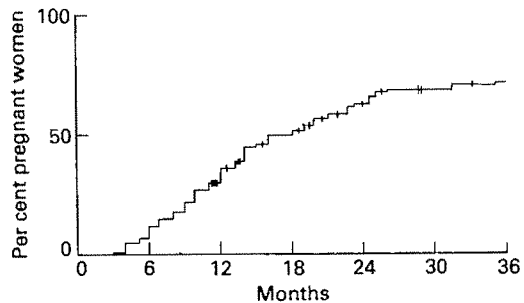
Data analysis

The cumulative proportion of women who became pregnant and were delivered of a live infant was calculated by the actuarial method and the curves obtained were compared by the log-rank test (Peto *et al.* 1977). The event data used in the computation of the probability to become pregnant and to give birth to a live child were the date of diagnosis (i.e. when all diagnostic procedures described in the previous section had been performed), the date of the last period, and the date of delivery.

The odds ratios (as estimators of relative risks) of a future miscarriage were examined in relation to the number of previous miscarriages, the age and socio-economic status of the women; 95% confidence intervals were calculated (Miettinen 1976). In the computation of relative risks their potential reciprocal confounding effect was controlled for using stratification and the Mantel-Haenszel procedure (Mantel & Haenszel 1959). The trend in risk was assessed using the test described by Mantel (1963).

Results

The 3-year actuarial overall pregnancy rate was 72% (this analysis allows for the eight women who chose not to become pregnant) (Fig. 1). After 3 years, 64% of the women had had a

**Fig. 1.** Overall 3-year probability of becoming pregnant in 95 cases of repeated miscarriage, Milan, Italy, 1980–1986.

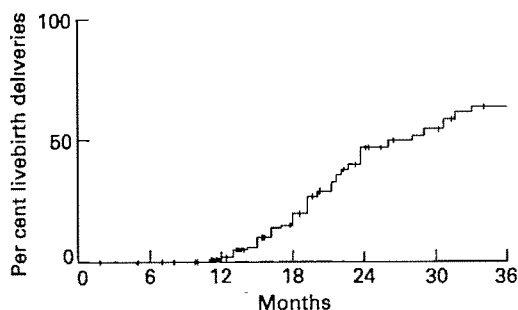


Fig. 2. Overall 3-year probability of a livebirth in 95 cases of repeated miscarriage, Milan, Italy, 1980-1986

livebirth, the frequency of term pregnancy increasing directly with time (Fig. 2).

The reproductive success rate decreased with the number of previous miscarriages, the rate was 80% after two, 60% after three and 46% after four or more miscarriages (χ^2 for trend adjusted for age = 4.76, $P = 0.03$, Fig. 3).

Women who were ≥ 30 years of age had a lower success rate than those who were younger, but this difference emerged only after 24 months and was not statistically significant (χ^2 for trend, adjusted for the number of previous miscarriages = 2.27, $P = 0.13$, Fig. 4). Socio-economic status was not associated with the probability of a livebirth. Similarly no difference in reproductive prognosis was observed according to the gestational age at the previous miscarriages (data not presented).

The prognostic role of previous miscarriages was analysed in the 66 observed pregnancies. Women with three miscarriages and those with

four or more had an increased risk of miscarriage (2.3 and 5.0 respectively) compared with those who had only had two previous miscarriages (χ^2 for trend adjusted for age = 5.2, $P = 0.02$, Table 3). There was no association with age or socio-economic status (data not presented).

Discussion

The major conclusions from this study are that the actuarial overall probability of a livebirth over 3 years in women with no livebirths and with two or more previous miscarriages is 64%, and the rate decreases with the number of previous miscarriages. This is a cautious estimate as we included in the analysis eight women who did not try to conceive during the study period. Bias should not be a problem in this study. The method of investigating the women with recurrent miscarriage and the clinical management did not change during the study period and all the women observed were considered. The fact that we studied only women with no previous successful pregnancy must have reduced the number of the unknown aetiological factors that were operating in our study group but it is likely that they consisted of a mixture of women, some with a cause for their repeated miscarriages and others who were just unlucky and had two or more miscarriages due to various factors (Gladden 1986; Warburton & Fraser 1961).

The proportion of women in our series in whom we could detect no cause for their miscarriages was 60% (95/158). This is comparable to,

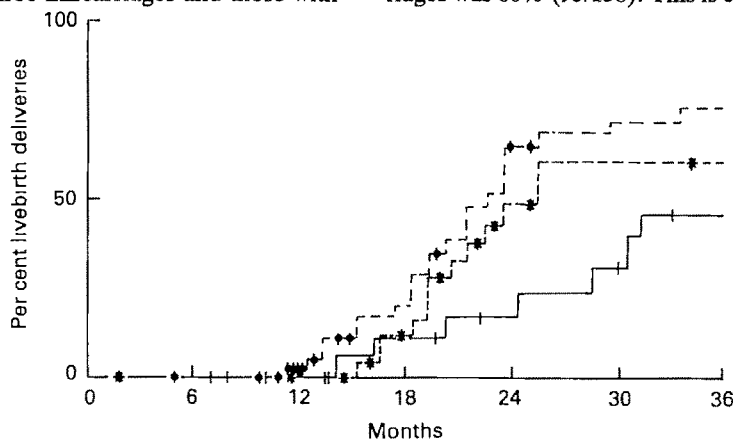


Fig. 3. Comparative 3-year probability of a livebirth according to number of previous miscarriages, Milan, Italy, 1980-1986. ---, Two previous miscarriages ($n = 43$), - · -, three previous miscarriages ($n = 29$), —, four previous miscarriages ($n = 23$).

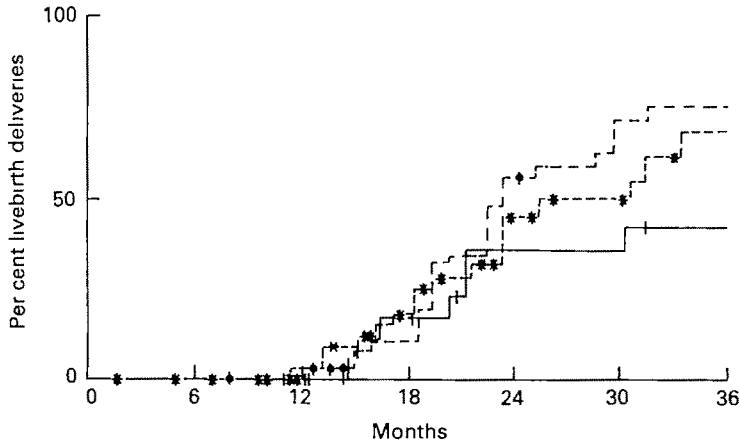


Fig. 4. Comparative 3-year probability of a livebirth according to age, Milan, Italy, 1980-1986. ---, ≤ 29 years old ($n = 31$); —, 30-34 years old ($n = 41$); —•—, ≥ 35 years old ($n = 23$)

although slightly higher than, that found by previous workers (Tho *et al.* 1979).

The use of beta-mimetics or cerclage should not inflate these estimates, in fact they were used respectively only in 23% and 3% of the women and according to clinical criteria similar to those used in previous pregnancy failures. Our relatively high livebirth rate is in general agreement with other recent series. Stray-Pedersen & Stray-Pedersen (1984) in Oslo reported 61 pregnancies and 40 livebirths in 85 women with recurrent miscarriages of unknown aetiology; similarly, in North America, Harger *et al.* (1983) studied 41 women with recurrent miscarriages of unknown aetiology and observed 31 pregnancies and 24 livebirths. However, in both these studies there is little information on the length of follow-up.

Table 3. Outcome of 66 observed pregnancies according to number of previous miscarriages in 95 patients with recurrent miscarriage, Milan, Italy, 1980-1986*

Number of previous miscarriages	Outcome of pregnancy		Odds ratio† (95% confidence interval)
	Miscarriage	Livebirth	
2	5	25	1‡
3	7	15	2.3 (0.6-8.5)
≥ 4	7	7	5.0 (1.2-20.1)

χ^2 for trend, adjusted for age = 5.2, $P = 0.02$.

* Twenty-nine women did not conceive.

† Adjusted for age.

‡ Reference category.

The number of previous miscarriages is the major determinant of reproductive prognosis. The probability of a livebirth in the next 3 years was 35% lower in women with four or more previous miscarriages than in those who had two previous miscarriages. This may be partly explained by the number of chance miscarriages in the subgroups, perhaps decreasing with the number of previous failures. Similarly, the risk of a subsequent miscarriage rose with the number of previous miscarriages. Age and socio-economic status, an indirect index of attention to health care, did not influence the results.

The study was limited to 3 years to obtain sufficient numbers for proper analysis. However, five of the women were observed for 6 years and four of them had had a livebirth by that time. The facts that the proportion of livebirths increases with time, and the favourable reproductive prognosis in women who have no known cause for their history of repeated miscarriages, suggest the importance of only using controlled clinical trials to test the effect of any new treatment that might prevent recurrent miscarriage.

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Occult biochemical pregnancy: fact or fiction?

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Summary. Daily urine samples were obtained from 38 women of proven fertility, who were attempting to conceive, to assess the frequency of clinically unsuspected 'biochemical pregnancies' on the basis of late luteal phase rises of hCG. In 25 clinically confirmed conception cycles, the earliest pregnancy-induced hCG rises from baseline occurred 8 days after the endocrinologically defined date of ovulation (mean 11.1 days, SD 1.3). Urine samples obtained throughout 50 non-conception ovulatory cycles provided no evidence of any 'biochemical pregnancies' on the basis of late luteal phase rises of hCG occurring 8 or more days after the endocrinologically defined date of ovulation. If, as in previous studies, the date of ovulation had been estimated on the basis of menstrual cycle length and only the relatively non-specific NIH anti-serum had been used in RIAs, 7 of the 50 (14%) cycles would have been classified as resulting in biochemical pregnancies on the basis of hCG >50 m-i.u./ml on one occasion or >20 m-i.u./ml on two or more occasions after day 20 of the cycle. These levels of 'hCG' occurred at or soon after ovulation and could be explained by LH cross-reacting in the RIA. The rises of 'hCG' were not confirmed when the urine samples were retested with an antiserum (Wellcome 895) of very high specificity for hCG. These results raise the possibility that the frequency of clinically unsuspected 'biochemical pregnancy' may have been substantially overestimated.

Several studies have been published suggesting that many women miscarry pregnancies at such an early stage that neither they nor their attendants recognize that a pregnancy has occurred. In most studies the subjects have been women attending infertility clinics, and samples have been obtained following stimulation of ovarian function with or without in-vitro fertilization

(IVF) procedures. Few comparable data on the normally fertile woman have been published. The evidence for a pregnancy having occurred and then failed in two such studies was based on the detection of a transient rise in human chorionic gonadotrophin (hCG) in either urine samples obtained on day 21 of the menstrual cycle and alternate days thereafter until menses (Miller *et al.* 1980; Edmonds *et al.* 1982) or a single serum sample obtained 'about day 25' in the luteal phase (Whittacker *et al.* 1983). The quoted incidence of these 'biochemical pregnancies' in women trying to conceive varies from 8% to 45.6% of cycles. The timing of ovulation was not known in these studies but estimated on the basis of cycle length. Luteinizing hormone (LH) and hCG have very similar molecular structures,

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and some degree of cross-reaction occurs in many radioimmunoassays. Without definite knowledge of the timing of ovulation it is not possible to exclude the possibility that transient rises in hCG after day 20 of the menstrual cycle were due to cross-reaction with late ovulatory surges of LH.

Our study examined the frequency of early pregnancy failure with particular reference to collection of daily urine samples from day 5 of the cycle until menses, in normal fertile women, in order to establish the date of ovulation using assays for LH and ovarian steroid metabolites, and the use of antisera of low (NIH SB6) and high (Wellcome 895), specificity for the estimation of hCG excretion.

Patients and methods

Forty-two women volunteered to participate in the study. Two women failed to collect samples according to protocol and were excluded. Two women failed to conceive within 6 months and were excluded on the grounds of potential sub-fertility. The remaining study group of 38 women all conceived a clinical pregnancy within 6 months. Of the study group of 38 volunteers, 18 were recruited from the family planning clinic having had an intrauterine contraceptive device removed with a view to attempting to conceive, and a further four were recruited having previously used sheaths for contraception. Sixteen women were recruited from the obstetric wards, having had a first trimester miscarriage and were asked to use sheaths until their next normal period before trying to conceive again. The mean age of the study group women was 27.4 years (range 22–38 years). Thirty-six women had had at least one previous pregnancy and two had never conceived before. Women with a past history of infertility were not invited to enter the study. A detailed menstrual history was taken by one of us (E.W.) from each woman with particular reference to normal cycle length.

The volunteers were asked to collect 20 ml of an early morning specimen of urine each day from day 5 of their cycle until either their next menstrual period began or until pregnancy was confirmed. The urine was collected in bottles, labelled with name, date and day of cycle and stored frozen until collected for assay. All the women entering the study were attempting to conceive and were aware of the ideal timing of intercourse for conception and none had hus-

bands whose work took them away from home or involved shift work which would interfere with intercourse during the fertile phase.

Urinary hormone assays

Urine samples were radioimmunoassayed for LH and expressed as i.u./g creatinine using a universal liquid phase immunoassay with iodinated tracer obtained from Mr K. Ferguson, Department of Biochemical Endocrinology, Chelsea Hospital for Women. The method employed a rabbit antiserum raised against human LH (WRB/F87) supplied by Professor W. R. Butt. A sample of the first IRP for LH (68/40) designated as containing 77 i.u./ampoule was obtained from NIBSC. Urine samples were radioimmunoassayed for oestrone 3-glucuronide (Baker *et al.* 1979) and pregnanediol 3-glucuronide (Samarajewa *et al.* 1979) and results expressed as μg , and mg/g creatinine respectively.

Radioimmunoassays for hCG used two rabbit antisera (SB6 from the National Institutes of Health, final dilution 1:72 000, and Wellcome Diagnostics antiserum no. 895, final dilution 1:24 000). Brief details of the hCG RIA protocol are as follows: 200 μl normal male urine (standard curve) or 200 μl urine sample, 200 μl standard hCG (2nd IRP) diluted in 0.01 M phosphate-buffered saline (PBS), pH 7.8, supplemented with 0.5% BSA or 200 μl buffer, 100 μl anti-hCG diluted in buffer supplemented with 0.01 M EDTA, and 100 μl ^{125}I -labelled hCG (specific activity 80–100 $\mu\text{Ci}/\mu\text{g}$) were added in sequence to 12 $\times$ 75 mm polystyrene tubes. After 20 h at 22°C the assay was separated using 200 μl precipitating antibody, donkey anti-rabbit from the Scottish Antibody Production Unit (SAPU), Law Hospital, Carlisle, Lanarkshire, and 750 μl 10% PEG 6000, diluted in PBS/EDTA and PBS/BSA buffers respectively. We determined the cross-reactivity of LH (1st IRP, 68/40) in the SB6 assay as 6% and in the Wellcome 895 assay as 0.25%. Reproducibility of the assays, assessed as coefficients of variation, was similar, 6 to 9% (intra-assay) between 3.0 and 31 m-i.u./ml and less than 12% (inter-assay) up to 45 m-i.u./ml.

Definition of ovulation timing

Ovulation was defined as occurring the day of the first rise of LH during the LH peak when this rise was followed by at least a doubling of preg-

nanediol within 72 h (De Crespigny *et al.* 1981). A rise in LH was defined as a level at least three times the mean of the previous 3 days.

Results

A total of 79 cycles were studied, four of which were anovular. In the remaining 75 cycles a clinically confirmed pregnancy occurred in 25. Twenty-one of these pregnancies continued into the third trimester, and in four there was histological confirmation of first trimester abortion. Of the 13 women who contributed cycles but conceived after the study was completed, two pregnancies miscarried in the first trimester and 11 resulted in the birth of a live infant. In the remaining 50 cycles there was endocrinological evidence of ovulation and normal menstruation, but no clinical signs of pregnancy although all the women who contributed cycles did conceive within 6 months of abandoning contraception.

Timing of ovulation

In the 75 ovulatory cycles, ovulation, as defined endocrinologically, occurred within the range of 8–31 days after the first day of the last menstrual period (mean 16.4 days, SD 4.0). In 21 cycles ovulation occurred on or after day 18 of the cycle and in only 11 of these would late ovulation have been suspected on the basis of the menstrual history stating cycles of greater than 32 days.

Duration of the mid-cycle LH surge and cross-reaction of LH in assays for hCG

In the 75 ovulatory cycles the mid-cycle rise in urinary LH lasted for between 2 and 7 days (mean 4.9 days, SD 1.4) before returning to baseline.

Using the SB6 antibody in the hCG RIA we found elevations of urinary 'hCG' above baseline in 74 of the 75 ovulatory cycles during the days of the mid-cycle urinary LH rise. These could be attributed to the cross-reaction of LH. The range of 'hCG' levels during the LH surge was 4.3 to 101 m-i.u./ml, mean 27.2 (SD 23.3). These elevations of 'hCG' lasted for between 1 and 7 days (mean 3.3, SD 1.5). In 26 of these cycles the specimens were retested with antisera of greater specificity (Wellcome 895) and in all cycles tested the measured levels of 'hCG' were substantially reduced. Only 15 of the 26 cycles showed any rise above the 'hCG' baseline during

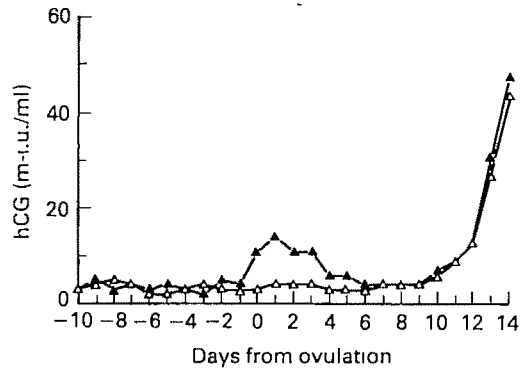


Fig. 1. Comparison of urinary hCG (mean+SE) in 21 conception cycles using SB6 (▲) and Wellcome 895 (△) antisera.

the LH surge using the Wellcome 895 antibody. Of these 15, the mean length of elevation was 1.9 days (range 1–4 days).

hCG during conception cycles

In the 25 conception cycles, the earliest evidence of pregnancy on the basis of hCG levels elevated above baseline (measured with the SB6 antiserum) was 8 days (mean 11.1 days, SD 1.3, range 8–13 days after ovulation). In 21 of the 25 conception cycles hCG was measured with both SB6 and Wellcome 895 antisera and the data obtained are shown in Fig. 1. These pregnancy-induced rises of hCG were recognized equally well using the NIH SB6 and the Wellcome 895 antibodies. The mid-cycle rise of hCG detected using SB6 (due to cross-reaction of LH) was greatly reduced with the Wellcome 895 antibody.

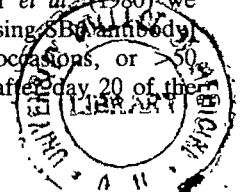
hCG in 50 ovulatory non-conception cycles

Between the 8th day after ovulation and subsequent menstruation there were no elevations of hCG above baseline suggestive of 'occult biochemical pregnancy'. In Fig. 2 the mean values and ranges of hCG (using SB6 antibody) throughout these cycles are shown. No hCG values above 20 m-i.u./ml were detected 7 or more days after ovulation.

hCG after day 20 of the menstrual cycle

Using the criteria of Miller *et al.* (1980) we detected hCG elevations (using SB6 antibody) of >20 m-i.u./ml on two occasions, or >50 m-i.u./ml on one occasion after day 20 of the

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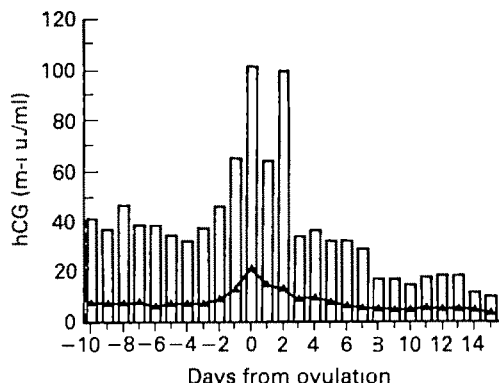


Fig. 2. Mean and range of urinary hCG using SB6 antibody in 50 non-conception cycles showing absence of levels above 20 m-i.u./ml after day 7 following ovulation.

cycle, in nine of the 50 non-conception cycles. Two of the nine cycles would have been expected to have late LH surges on account of long cycle length and would be excluded by Miller's criteria which estimated date of ovulation on this basis. In these two cycles, as well as in the other seven in which high 'hCG' values were detected using the SB6 antiserum after day 20, the rises of hCG coincided with the LH surge. It was possible to retest for hCG in five of these cycles using Wellcome 895 antibody, and in no case was a mid-cycle rise of hCG detected. The mean values for 'hCG' in these five cycles using the two antisera are shown in Fig. 3.

Discussion

This study has shown that when looking for evidence from urine of occult biochemical pregnancies, it is essential to define the date of ovulation and the duration of the mid-cycle LH surge. This is especially important when the antiserum to hCG cross-reacts substantially with LH. In this study of normally fertile women, ovulation occurred after day 18 in 21 of 75 cycles, but in only 11 of these would late ovulation have been predicted on the basis of menstrual history. The mid-cycle elevation of LH in urine is more prolonged than in serum, ranging in this study from 2 to 7 days. These two factors led to the urinary LH surges lasting beyond day 21 in a substantial number of cycles. If a hCG antiserum is used which cross-reacts with LH there is a clear danger of overdiagnosing the incidence of 'occult biochemical pregnancy'.

Previous studies of the frequency of 'occult biochemical pregnancy' used menstrual history

to define the date of ovulation and interpreted elevated hCG values on or after day 21 of the cycle as evidence of pregnancy. If the criteria used by Miller *et al.* (1980) in their large study had been applied to our data, we would have detected a comparable rate of 14% 'occult biochemical pregnancy'; but by defining the date of ovulation and the duration of the LH surge, the elevations of hCG detected using SB6 antiserum, could be more appropriately explained on the basis of cross-reaction with LH.

In the 25 conception cycles which we studied, the earliest evidence of a pregnancy-induced rise in hCG occurred 8 days after the onset of the LH surge. It would seem unlikely that non-viable conceptions would be detectable any earlier after ovulation by elevations of urinary hCG.

Edmonds *et al.* (1982) defined any urinary hCG value above 56 m-i.u./ml after day 21 of the cycle as indicative of pregnancy on the grounds that this was the upper limit of the range observed in a group of sterilized women. The date of ovulation was not known in these sterilized women and it is possible that the upper limit of the range was due to cross-reaction with LH. Miller *et al.* (1980) used the criteria of two or more values above 2 ng/ml (20 m-i.u./ml) or one or more of 5 ng/ml (50 m-i.u./ml) on or after day 20, but stated that these limits were chosen on empirical grounds. In our own study the upper limit of the range of hCG values detected with the SB6 antiserum during the follicular phase more than 3 days before ovulation was 46.7 m-i.u./ml. In the non-conception cycles, the upper limit of the range of hCG values detected with the SB6 antiserum in the late luteal phase (defined as the period from day 8 after ovulation

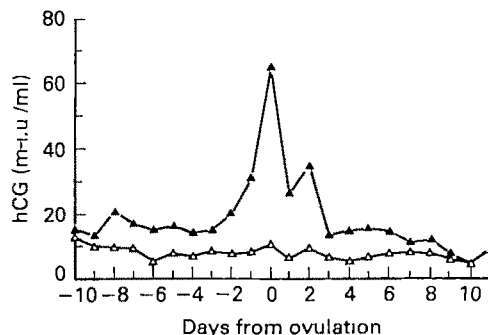


Fig. 3. Mean hCG levels centered round day of ovulation, in five non-conception cycles with hCG >20 m-i.u./ml after 21 of the cycle (using SB6 antiserum) showing lower levels with Wellcome 895. ▲, SB6; △, Wellcome 895.

to menstruation) was 18.3 m-i.u./ml, which was well below the limits which could be interpreted as indicative of pregnancy. During the LH surge 'hCG' concentrations considerably higher than 50 m-i.u./ml were frequently detected as can be seen from Figs 2 and 3, indicating that the per cent cross-reaction observed by testing LH reference preparations tested in the SB6 assay does not accurately reflect the degree of interaction of LH excreted during the surge with the SB6 antiserum.

Wilcox *et al.* (1985) came to similar conclusions as our own as to the infrequency of biochemical pregnancy in their study of 72 cycles. They used a highly sensitive IRMA assay for β -hCG together with the SB6 assay. Four cycles showed raised β -hCG on the IRMA assay, but two of these were early clinical miscarriages following periods of amenorrhoea and symptoms of early pregnancy. One cycle had raised β -hCG during menses and the fourth fulfilled the criteria of biochemical pregnancy of Miller *et al.* (1980) and Edmonds *et al.* (1982). Although it is possible that if we had had access to a more sensitive method of detecting hCG such as an IRMA we would have identified 'biochemical pregnancies' in our own study group, the data of Wilcox *et al.* (1985) do not support such a possibility.

Ours was a study of women with proven fertility, the majority of whom had been previously pregnant and all of whom conceived within a short period. It is possible that the incidence of 'occult biochemical pregnancy' is higher in women with unexplained infertility, undergoing ovulation induction or having in-vitro fertilization. To define accurately the frequency of biochemical pregnancy in these groups it will be necessary to define the date of ovulation and avoid the possibility of cross-reaction of LH in hCG assays. The problem of cross-reaction of LH can be considerably reduced by using antisera with greater specificity. The Wellcome antiserum (895) became available to us during the course of this study, and the false positive eleva-

tions detected using SB6 were eliminated with this antiserum which is highly specific for hCG. It has been shown that elevated hCG values in urine can occur sporadically in men and in sterilized women. Although we did not find any such elevations of hCG which could not be explained by LH cross-reaction or pregnancy, it is possible that this phenomenon could explain some of the 'biochemical pregnancies' which have been reported in previous studies, especially when few samples per cycle are tested. The evidence from our study indicates the potential for over-diagnosis of 'occult biochemical pregnancy' in cycles where the date of ovulation is unknown. It is possible that previous studies have substantially overestimated the frequency of this phenomenon.

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The changes in fetal habituation patterns due to a decrease in inspired maternal oxygen

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Summary. Habituation is widely regarded as a basic form of learning. A fully functioning central nervous system is probably required for a normal habituation pattern. Habituation of the behavioural response to vibroacoustic stimulation was studied in 23 normal human fetuses whose gestational age was 36 or more weeks. Eighteen were tested whilst their mothers breathed either air or a 12% oxygen in nitrogen mixture. The test was repeated the following day with the order of the gases reversed. In addition five fetuses were tested while their mothers breathed air on both occasions. Of the 18 fetuses 17 had a normal habituation pattern when tested in air but only two had a normal pattern when their mothers breathed 12% O₂. Four of the five fetuses tested both days in air had normal patterns on the second day. Altering the amount of inspired maternal O₂ alters the fetal habituation pattern.

Habituation is the decrease in responsiveness that occurs when an organism is repeatedly stimulated. This is widely regarded as a form of learning (Buchwald & Humphrey 1973; Stevensen & Siddle 1983) and there is good evidence that a normal habituation pattern reflects an intact and fully functioning central nervous system (Lewis 1971; Leader *et al.* 1984; Madison *et al.* 1986*a,b*).

Fetal habituation to repeated vibroacoustic stimulation has been demonstrated in the human fetus (Leader *et al.* 1982*a,b*; Birnholz & Benacerraf 1983; Madison *et al.* 1986*a*). Fetuses who had decreased growth velocity as measured by serial ultrasonic estimations, meconium staining of the amniotic fluid, or who were born small for gestational age, had habituation patterns that differed significantly from infants born to mothers in optimal obstetrical circumstances (Leader *et al.* 1982*a*).

Baillie (1974) described the antenatal oxygen stress test and measured the maternal ventilatory, blood gas and cardiovascular variables as well as the fetal cardiovascular responses to an inspired maternal oxygen concentration of 11.6%. He showed that this oxygen concentration was safe to use and that the critical discriminant factor was the time taken for the heart rate to return to its baseline value, if the fetus developed a tachycardia in response to this hypoxic stress.

As this test was in routine clinical use in high-risk cases at Groote Schuur Hospital, a study was undertaken to assess the effect of 12% ($\pm 0.5\%$) oxygen on antenatal fetal habituation patterns. This study differed from the standard oxygen stress test in that the mothers were not given an injection of pethidine to induce a steady state before the test.

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Patients and methods

Twenty-three women who were 36 or more weeks pregnant, did not smoke, and had an uncomplicated antenatal course, were recruited for the study. The aims and the protocol were explained

and informed consent was obtained. Subjects were randomly allocated to one of three groups.

Group 1

The fetuses of 10 subjects were initially tested for habituation whilst breathing room air. The test was repeated at approximately the same time the following day while the subjects were breathing 12% oxygen.

Group 2

Eight fetuses were initially tested while their mothers were breathing 12% oxygen and the test repeated at approximately the same time the next day while their mothers breathed room air.

Group 3

Five fetuses were tested when their mothers were breathing room air and were then retested the following day while their mothers were breathing air through the same apparatus used to deliver the 12% oxygen mixture. Subjects were not informed that the gas mixture they were breathing was any different to that used in the other two groups.

Stimulus

A standard Ronson electric toothbrush motor was once again used (Leader *et al.* 1982a). In all cases fetal movements were measured simultaneously by three independent methods: a modified Hewlett-Packard A8021A cardiotocograph (Leader & Baillie 1979), a linear-array real-time ultrasound scanner (Aloka, Axiscan), and maternal observation of fetal movements.

The women were placed in the left lateral position on a soft couch; they were shown the toothbrush, the procedure explained and the stimulus demonstrated. Fetal movements were recorded by an independent observer (E.V.) watching the real-time ultrasound image. The toothbrush was placed on the maternal abdominal wall over the fetal head and a 5-s stimulus applied approximately every 20 s.

Behavioural response

Fetal movements in response to the stimulus were measured simultaneously by all three methods. Only movement that occurred during the stimulus or within 2.5 s of its cessation was considered a response. Movement occurring after that time was considered spontaneous. If spontaneous fetal movement was present when the stimulus was due, it was withheld until the

movement ceased. Lack of a response to five consecutive stimuli indicated habituation and the number of stimuli required to produce habituation was recorded. These criteria are similar to previous studies (Leader *et al.* 1982a; Madison *et al.* 1986a).

The fetal heart rate was monitored using the cardiotocograph during the period preceding stimulation and again immediately afterwards. It was technically difficult to monitor the fetal heart rate during stimulation due to interference between the real-time ultrasound and the Doppler used for recording the heart rate. It was therefore only possible to observe the fetal heart rate intermittently using the ultrasound transducer.

Oxygen mixture

The specially prepared mixture of 12% ($\pm 0.5\%$) oxygen in nitrogen was breathed by the woman through a non-return Ambu valve using a flow rate of 25 l/min to prevent any rebreathing and carbon dioxide accumulation. A physiological mouthpiece and nose clip precluded air breathing (Baillie 1974). Subjects were initially started on room air via the same apparatus to allay any anxiety. They then breathed the 12% O₂ for 15 min before they were tested for habituation and continued on it throughout the entire period of stimulation.

Habituation patterns

Using the same experimental protocol, a previous study examined the number of stimuli required to produce habituation in 40 subjects who had a normal uncomplicated antenatal, intrapartum and neonatal course and who were delivered of infants thought to be in an optimal condition. All fetuses were tested 10 days or less before delivery. Of the 40 fetuses 37 habituated after 10–50 stimuli had been given. The remaining three were still responding after 50 stimuli (Leader *et al.* 1982a).

A further study (Leader *et al.* 1982b) examined the habituation patterns of 46 fetuses who were born small for gestational age, 28 who had meconium staining of the amniotic fluid and 38 in whom serial ultrasound measurements of the fetal biparietal diameter showed a lag in the expected rate of growth. Compared with the normal group, the high-risk fetuses showed three different patterns described previously (Patterson & Venables 1978): non-responders, fast habituators (requiring one to nine stimuli) and

slow habituators (requiring >50 stimuli). These patterns differed significantly from the optimal group of fetuses and have been widely found in a variety of central nervous system (CNS) pathologies (Leader *et al.* 1982b).

When tested at 1 year of age using the Griffiths Mental Developmental Scale, infants who habituated to between 10 and 50 stimuli were found to

Table 1. Habituation results in 10 subjects who breathed air followed by 12% O₂ the next day

Subject no.	Number of stimuli for habituation	
	Day 1 Air	Day 2 12% O ₂
200	25	>51*
201	15	6
203	13	49†
205	20	>51
400	14	1
403	19	1
408	22	>51
410	29	>51
412	34	>51
414	24	>51

* >51 stimuli indicates that the fetus failed to habituate by 51 stimuli.

† This fetus was found to have meconium staining of the amniotic fluid when labour started 3 days later.

Table 2. Habituation results in eight subjects tested while breathing 12% O₂ first and air the next day

Subject no.	Number of stimuli for habituation	
	Day 1 12% O ₂	Day 2 Air
199	>51*	19
207	>51	2†
238	>51	16
250	18	26‡
402	>51	39
404	>51	29
406	>51	20
409	>51	22

* >51 stimuli indicates that the fetus failed to habituate by 51 stimuli.

† This fetus had an arterial base deficit of 17 meq at caesarean section; the infant was small for gestational age; Apgar scores were 9 and 10 at 1 and 5 min respectively.

‡ This fetus was found to have thickly meconium-stained amniotic fluid when labour started 4 days later.

have a significantly better performance (Leader *et al.* 1984).

Habituation to between 10 and 50 stimuli was regarded as a normal pattern. Fetuses habituating to between one and nine stimuli were labelled fast habituators (FH) and those still responding after 51 stimuli slow habituators (SH) and regarded as abnormal.

Results

The fetuses of all 10 subjects in group 1 had normal habituation patterns when tested while their mothers were breathing air. When repeated the next day breathing 12% O₂, only one had a normal habituation pattern (Table 1). Seven of the eight fetuses in group 2 tested initially while their mothers breathed 12% O₂ had an abnormal habituation pattern. When tested the following day in air, seven of the eight had a normal habituation pattern (Table 2).

In the control group, all five fetuses had a normal habituation pattern when tested in air. When tested the next day while their mothers breathed air through the same apparatus, four of the five had a normal pattern (Table 3). Subject no. 413 failed to habituate by 51 stimuli. This fetus was found to have meconium-stained amniotic fluid in labour 5 days later.

Of the 18 fetuses tested in air 17 had a normal habituation pattern. When tested while their mother breathed 12% O₂ only two had a normal pattern ($P < 0.001$, χ^2 with Yates' correction).

The fetal heart rate showed no abnormal changes during the period of reduced inspired maternal oxygen. Fetal stimulation was usually associated with an increase in the heart rate.

Discussion

This study shows quite clearly that reduction of the oxygen concentration inspired by the mother to 12% alters the fetal habituation pattern. Seventeen of the 18 fetuses had a normal pattern when tested in air but only two had a normal pattern when tested in 12% O₂. The commonest abnormal pattern, seen in 13 fetuses, was the failure to habituate by 51 stimuli. Three of the four fetuses, whose habituation patterns were at variance to the rest of the group, were found to have meconium staining of the amniotic fluid when labour started 3–5 days later. The remaining fetus, who had an abnormal pattern when tested in air, was found to have an arterial base deficit of 17 meq at caesarean section performed for fetal distress and was found to be small for

Table 3. Habituation results in five subjects tested initially in air followed the next day by air through the same apparatus as the 12% O₂ (subjects were unaware which gas mixture they were breathing)

Subject no.	Number of stimuli for habituation	
	Day 1 Air	Day 2 Air
398	22	18
401	33	16
405	23	10
407	11	10
413	32	>51*†

* >51 stimuli indicates that the fetus failed to habituate after 51 stimuli.

† This fetus was found to have thickly meconium-stained amniotic fluid when labour started 5 days later.

gestational age. A high prevalence of abnormal fetal habituation patterns is associated with small-for-gestational-age infants and meconium-stained amniotic fluid (Leader *et al.* 1982b).

Studies in fetal sheep (Natale *et al.* 1981; Leader *et al.* 1986) have shown decreased fetal forelimb activity during periods of fetal hypoxia produced by reducing the inspired oxygen concentration to the ewe. In addition, it was found that fetal sheep failed to show either fetal movement, blood pressure or heart rate responses to cutaneous stimulation during periods of hypoxia (Leader *et al.* 1986). If they did respond, they were found to habituate more rapidly than controls. The differences in the effect of hypoxia between the sheep and human studies are probably due to the greater degree of hypoxia induced in sheep.

Breathing 12% O₂ is the equivalent of living at 13 000 feet above sea level and this reduced the maternal PaO₂ from 99 mmHg (SD 4.3) to 44 mmHg (SD 2.8). The arterial O₂ saturation falls to 86% and has been shown by Baillie (1974) to be safe. Wood *et al.* (1971) did report adverse fetal responses in mothers who were breathing 10% O₂ in labour.

Previous studies have examined the fetal cardiovascular responses during inspired O₂ using concentrations of 8–20% (Hellman *et al.* 1961; John 1965; Copher & Huber 1967). The mild tachycardia observed in some of these studies may have been induced by maternal anxiety and catecholamine release. In the present study, much time was spent allaying maternal

apprehension by allowing mothers to become used to the breathing apparatus before altering the amount of oxygen. Four of the five fetuses in the control group who breathed air through the same apparatus had a normal habituation pattern.

In animals, hypoxia is associated with a rise in fetal catecholamines (Jones & Robinson 1975; Lewis *et al.* 1984). In the present study it is unlikely that changes seen in the habituation patterns were due to increased fetal catecholamine release as the degree of hypoxia induced in animals is far greater. In addition, when tested during an intravenous infusion of noradrenaline, fetal sheep habituate more rapidly compared with controls (four stimuli compared with 14, $P < 0.001$, Leader *et al.* 1986). In the present study, the most common habituation pattern during hypoxia was a failure to habituate by 50 stimuli. This is not in keeping with the effects of catecholamines.

Some studies have shown that hypoxia may alter the fetal electrocortical (ECoG) activity (Harding *et al.* 1981; Clewlow *et al.* 1983; Natale *et al.* 1981). It is unlikely that effects described in this study are due to changes in fetal state as the reduction in the oxygen saturation produced by the O₂ concentration used is very small (Baillie 1974).

In addition, a study using a broad spectrum vibroacoustic stimuli in fetal sheep showed that fetal movement responses and habituation were independent of fetal ECoG activity at the time of stimulation (Leader *et al.* 1988).

In fetal primates and sheep, there is a considerable increase in the fetal cerebral blood flow during initial periods of hypoxia (Purves & James 1969; Behrman *et al.* 1970; Tweed *et al.* 1983). Different areas within the fetal brain show independent changes in blood flow. With increasing severity of hypoxia, the flow to the cerebral cortex is the first to show a decrease during hypoxia whilst the brain stem is the last.

If both the fetal movement response to stimulation and habituation are mediated via cortical pathways, it may explain why the alterations in habituation patterns were so consistent during 12% O₂ compared to the earlier studies on heart rate changes in which quite inconsistent responses were obtained.

The results of this study provide further evidence that fetal habituation may provide a more sensitive measure of central nervous system integrity.

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Gestational-age-dependent concentrations of polyamines, their conjugates and metabolites in urine and amniotic fluid

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Summary. The metabolism and fate of polyamines during cell proliferation, differentiation, and cell loss were investigated by measuring the concentration of polyamines, their conjugates and some of their metabolites in amniotic fluid of 24 subjects, and in urine of 85 women during pregnancy. The increase of putrescine and spermine in acid-hydrolysed urines during pregnancy appeared to be almost completely due to increases in monoacetylated putrescine and N^1, N^{12} diacetylated spermine, respectively. The latter two were the quantitatively most important polyamines in amniotic fluid. In urine, monoacetylated putrescine showed the highest levels at the end of pregnancy, whereas N^1, N^{12} diacetylated spermine reached the highest values at about 32 weeks gestation. It was impossible to establish whether extracellular monoacetylated putrescine is linked either to cell growth or cell loss. The appearance of N^1, N^{12} diacetylated spermine is probably due to cell loss and dependent on the degree of differentiation during fetal development. The decline and eventual disappearance of urinary N^1, N^{12} diacetylated spermine during the first 2 years after birth may be coherent with maturation of the FAD-dependent polyamine oxidase activity.

The polyamines putrescine, spermidine and spermine, are a group of low molecular weight cations that are ubiquitous in nature. Most probably by their association with nucleic acids, they are indissolubly linked with growth processes (Pegg 1986). The correlation of elevated intracellular polyamine synthesis with rapid growth is

well established. Increased extracellular polyamine levels have been linked either to cell growth (putrescine) or to cell loss (putrescine and spermidine) (Russell 1983). Increased extracellular polyamine concentrations are found in patients with a variety of neoplastic diseases (Russell 1983).

Since the development of the feto-placental unit is characterized by both rapid growth and high naturally occurring cell loss, the study of pregnancy may increase our understanding of neoplastic diseases.

Embryonic and placental waste products appear in the fetal and maternal circulation and in amniotic fluid, and are finally excreted in the maternal urine. During pregnancy, significant elevations of putrescine and spermine have been observed in maternal plasma and acid-hydro-

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lysed urine (Russell *et al.* 1978; Muskiet *et al.* 1984; Hiramatsu *et al.* 1985). The levels normalize within the first days postpartum. Large amounts of amine oxidases are present in placenta (Bradsley *et al.* 1974; Illei & Morgan 1979; Gahl & Pitot 1981a) and amniotic fluid (Tufveson 1978; Gahl & Pitot 1981a). During gestation the activity of polyamine-degrading enzymes, notably amine oxidases, increases in maternal blood (Carrington *et al.* 1972; Morgan 1982; Gahl *et al.* 1982; Morgan *et al.* 1983).

The first and rate-limiting step in the degradation of polyamines is *N*-acetylation. The acetyl conjugates are rapidly degraded by amine oxidases (Seiler *et al.* 1981; Pegg 1986). Mammalian copper-containing amine oxidases catalyse the oxidative deamination of the terminal amino groups of both parent and acetylated polyamines to yield products that cannot be reconverted into polyamines ('terminal catabolism') (Seiler *et al.* 1985). Polyamine oxidases (FAD dependent) catalyse the oxidative cleavage of acetylated spermidine and spermine, which are split at the secondary amino groups, producing putrescine from *N*¹-acetylspermidine, and spermidine from *N*¹-acetylspermine (interconversion) (Seiler *et al.* 1985).

In an effort to define more clearly the metabolism and fate of the polyamines during alterations in cell proliferation, differentiation, and cell loss, we investigated the levels of polyamines, their conjugates and some of their metabolites in amniotic fluid and urine during pregnancy.

List of non-standard abbreviations

DAP	1,3-diaminopropane
Pu	putrescine
Cad	cadaverine
Sd	spermidine
Sp	spermine
acDAP	monoacetyl-1,3-diaminopropane
acPu	monoacetylputrescine
acCad	monoacetylcadaverine
<i>N</i> ¹ acSd	<i>N</i> ¹ -acetylspermidine
<i>N</i> ⁸ acSd	<i>N</i> ⁸ -acetylspermidine
<i>N</i> ¹ acSp	<i>N</i> ¹ -acetylspermine
<i>N</i> ¹ , <i>N</i> ¹² diacSp	<i>N</i> ¹ , <i>N</i> ¹² -diacetylspermine
Isoputr	isoputrescine
Isoga	<i>N</i> -(3-aminopropyl)pyrrolidine-2-one
Putr	putrescine.

Patients and methods

Collection of urine and amniotic fluid

Early morning urine was collected daily from a

35-year-old woman from the 5th week of gestation until 2 weeks after delivery. Total 24-h urine specimens were collected from 85 pregnant women (mean age 28.8, SD 6.0 years) at various weeks throughout gestation and from 30 children, aged between 3 days and 15 years. All women had normal pregnancies, resulting in the birth of normal healthy babies. The specimens were acidified without delay to pH 1–2, with hydrochloric acid. Aliquots were stored at –20°C until analysis. Creatinine was measured by a picric acid method.

Amniotic fluid was collected by amniocentesis (diagnostic purposes) or at term (healthy women). The indications for amniocentesis were karyotyping, estimation of fetal lung maturity and the duration of pregnancy. The samples finally selected for the measurements of polyamines were obtained from women with no abnormalities with respect to karyotype, lecithin/sphingomyelin ratio or concentrations of alpha-fetoprotein (AFP). All the women gave birth to normal healthy babies. Samples were stored at –20°C until analysed.

Polyamine analyses

Before treatment, the amniotic fluid samples were deproteinized by adding an equal volume of a solution containing 120 g/l sulfosalicylic acid, freezing at –20°C, thawing at room temperature, and centrifuging at 1000 *g* for 10 min. Six sections (about 100 mg each) were cut from a normal placenta and extracted after homogenization (Potter apparatus) in 4 ml of 0.3 M HClO₄ and subsequent centrifugation at 1000 *g* for 10 min.

Polyamines, their conjugates and metabolites were isolated from 1 ml of urine, 3 ml of amniotic fluid and 2 ml of placental extract by adsorption onto silica gel. After conversion to their *N*-heptafluorobutyl-methyl esters they were determined by gas chromatography with nitrogen-phosphorus detection as described previously (van den Berg *et al.* 1986a). We only considered the sum of Isoputr and Isoga, as with the method used it was impossible to establish whether Isoga is a normal urinary excretory product or whether it is formed as a derivatization artefact of Isoputr (van den Berg *et al.* 1986a).

Results

Gestational-age-dependent urinary excretion of polyamines

No differences with respect to normal urinary levels of nine non-pregnant women in the same age group (van den Berg *et al.* 1986a) were found during pregnancy for acDAP, DAP, Pu, Cad, acCad, N^1 acSd, N^8 acSd, Sp, N^1 acSp and the ratio of N^1 acSd and N^8 acSd. Those polyamines which differed significantly are summarized in Table 1. Concentrations are expressed as a ratio of that of creatinine. Urinary acPu levels gradually increased during pregnancy. The concentration of N^1,N^{12} diacSp, a compound not detectable in urine of men and non-pregnant women by the present technique, increased during pregnancy to reach highest values between the 25th and 36th week. The urinary level of Putr was elevated during pregnancy, whereas the sum of Isoputr and its γ -lactam form Isoga was decreased.

Figure 1 illustrates the results for acPu and N^1,N^{12} diacSp from Table 1, together with data obtained in a longitudinal study in one woman between 5 weeks gestation and up to 2 weeks after delivery. These data show that elevated acPu levels rapidly normalized within 3 days after delivery and N^1,N^{12} diacSp reached the highest level at about the 32nd week, and disappeared from urine within 10 days postpartum. During pregnancy about 86% of Sp was excreted as N^1,N^{12} diacSp. When compared with previously established reference values (van den Berg *et al.* 1986a) none of the other polyamines showed differences in the percentage of acetylation (ratio of acetylated polyamine to the sum of free and acetylated polyamine, times 100).

Polyamine concentrations in amniotic fluid during normal pregnancy

Levels of free and acetylated polyamines, and some of their metabolites, were determined in 24 samples of amniotic fluid, collected at various stages (16–40 weeks) throughout gestation. Table 2 shows the median concentrations and ranges (nmol/l) which are large. Table 2 also shows the acetylation percentages of the various polyamines together with the molar ratio of N^1 acSd and N^8 acSd.

Polyamines in a placenta and in urine of newborns

Free and acetylated polyamine concentrations were determined in six sections of a normal term placenta. The mean concentrations (SD) expressed as nmol/g wet tissue, were: DAP 9 (3), Pu 3 (2), Cad 3 (1), acPu 3 (1), Sd 35 (9), N^1 acSd 2 (1), Sp 362 (42), N^1 acSp 28 (5), N^1,N^{12} diacSp 2 (1).

In contrast to adults, newborn infants excrete substantial amounts of N^1,N^{12} diacSp during their first month after birth. The mean (SD) urinary concentration in 15 babies (aged 3 days to 4 months) was 0.30 (0.14) mmol/mol of creatinine (range: 0.15–0.63). At the age of about 2 years N^1,N^{12} diacSp was no longer detectable in urine of infants, using the present method.

Discussion

During pregnancy, the levels of Pu and Sp increase in acid-hydrolysed urines (Russell *et al.* 1978; Muskiet *et al.* 1984; Hiramatsu *et al.* 1985). These increases can almost completely be attributed to elevations of acPu and N^1,N^{12} diacSp levels, respectively. Our results for excretion of acPu and N^1,N^{12} diacSp are in agreement with those for total Pu and Sp (Hiramatsu *et al.* 1985). In contrast to Russell *et al.* (1978) no sharp elevations of polyamines were found between 10 and 16 weeks gestation (Fig. 1).

In normal subjects 50% of Isoputr is present in urine as *N*-(3-acetamidopropyl)pyrrolidin-2-one (the acetylated form of Isoputr- γ -lactam, abbreviated Acisoga) (van den Berg *et al.* 1986b). In a period of increased excretion of total Isoputr, patients with non-Hodgkin's lymphoma excrete an even greater part as Acisoga (van den Berg *et al.* 1986b). In addition, Isoputr in urines of patients with non-Hodgkin's lymphoma is present in the form of a yet unknown, acid hydrolysable, conjugate. As the total Isoputr levels are also increased during pregnancy (Muskiet *et al.* 1984), the decreased excretion of Isoputr + Isoga (Table 1) may indicate, analogous to non-Hodgkin's lymphoma, that a relatively greater part is present as Acisoga and the conjugate of a structure as yet unknown. The high levels of total Isoputr and Putr may harmonize with the high activity of (Cu-containing) amine oxidases in placenta and maternal serum (Gahl & Pitot 1981b; Gahl *et al.* 1982).

Table 1. Gestational-age-dependent urinary excretion of polyamine metabolites in 85 women

Metabolite	Gestational age (weeks)								Non-pregnant* (n = 9)
	≤12 (n = 9)	13-16 (n = 12)	17-20 (n = 9)	21-24 (n = 10)	25-28 (n = 14)	29-32 (n = 10)	33-36 (n = 12)	37-40 (n = 9)	
acPu	1.70 (0.34)	1.85 (0.34)	2.13 (0.97)	2.03 (0.31)	2.53 (0.56)	2.34 (0.37)	2.50 (0.92)	2.95 (1.05)	1.43 (0.44)
Isoputr + Isoga†	0.23 (0.10)	0.25 (0.16)	0.14 (0.07)	0.22 (0.13)	0.18 (0.12)	0.30 (0.10)	0.24 (0.11)	0.17 (0.13)	0.71 (0.22)
Putr	0.30 (0.04)	0.37 (0.05)	0.32 (0.08)	0.39 (0.06)	0.41 (0.07)	0.41 (0.07)	0.42 (0.07)	0.38 (0.10)	0.25 (0.05)
N ¹ ,N ¹² diacSp	0.14 (0.05)	0.27 (0.08)	0.33 (0.13)	0.39 (0.09)	0.74 (0.31)	0.74 (0.50)	0.72 (0.30)	0.50 (0.29)	ND

* Non-pregnant women aged between 26 and 41 years

† Sum of Isoputr and Isoga.

ND, Not detectable.

See text for list of abbreviations

Results are mean (SD) values of mmol/mol creatinine.

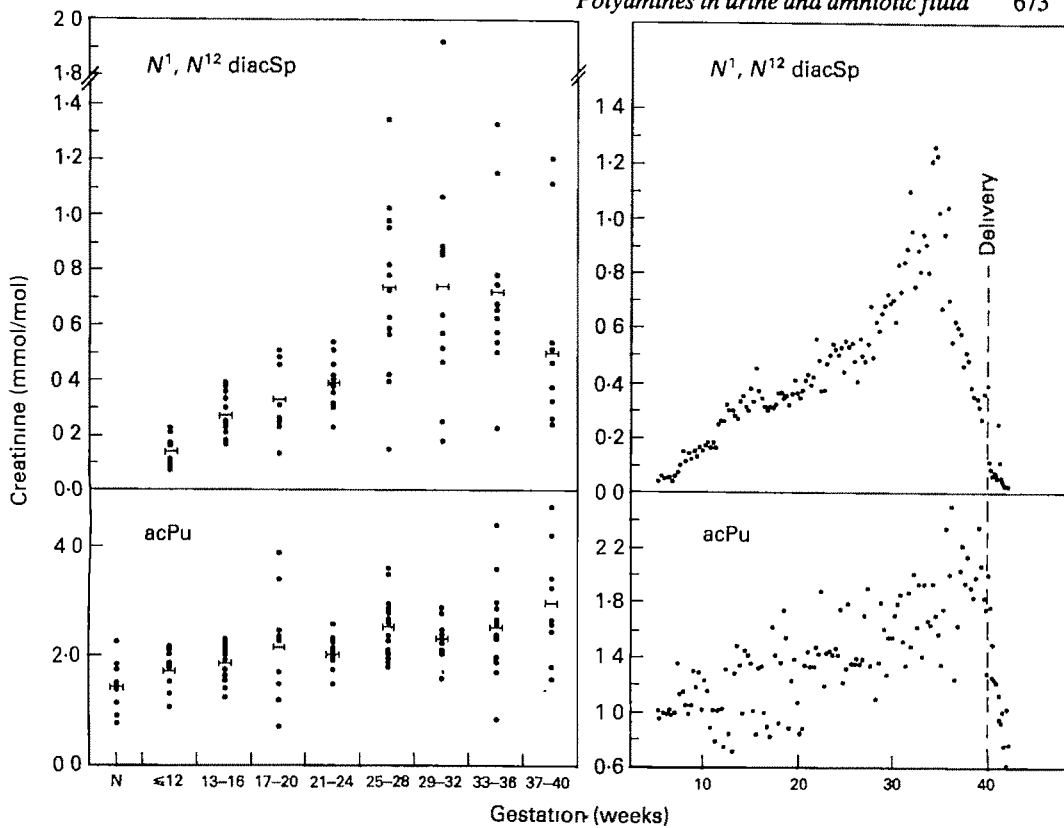


Fig. 1. Urinary levels of monoacetylated putrescine (acPu) and N^1, N^{12} diacetylated spermine (N^1, N^{12} diacSp) in 85 women (mean age 28.8, SD 6.0 years) at various weeks of the gestation period (left panel), together with the longitudinal study of a single subject from the 5th week of gestation up to 2 weeks after delivery (right panel). N, Level of nine non-pregnant women, aged 26-41 years; $\bar{x}$, mean value

The quantitatively most important polyamines in amniotic fluid are acPu and N^1, N^{12} diacSp. In contrast to urine (van den Berg *et al.* 1986a), all polyamines appear for the greater part as acetyl conjugates (see Table 2). In acid-hydrolysed amniotic fluid the Pu and Sd concentrations (Russell *et al.* 1978; Chan *et al.* 1979; Hiramatsu *et al.* 1985) are twice those calculated for the sum of acetylated and free Pu and Sd concentrations, respectively, in non-hydrolysed amniotic fluid. It is therefore likely that other forms of acid-hydrolysable conjugates of Pu and Sd, most probably peptide complexes (Chan *et al.* 1979), are present.

The most abundant polyamines in the placenta are free Sd and Sp. Their concentration is in good agreement with the literature (Gahl & Pitot 1981a). In contrast to other tissues (Pegg 1986), placental extracts appear to contain considerable amounts of monoacetylated polyamines and N^1, N^{12} diacSp. It is still unclear whether their origin is intra- or extracellular.

Alterations in extracellular putrescine levels

may reflect alterations in either cell growth or cell loss. Pu is presumed to originate from the removal of the acetylaminopropyl moiety from N^1 acSd by FAD-dependent amine oxidase (Pu as a 'degradation' marker), or its direct spontaneous leakage from growing cells (Pu as a 'growth' marker). Obviously, as Pu appears primarily as acPu in both urine and amniotic fluid, the free Pu, whatever its origins, is subsequently converted into acPu. The development of the fetoplacental unit is characterized by rapid growth. Most hormones which play a role in pregnancy are known inducers of ornithine decarboxylase, which produces Pu from ornithine (Jänne *et al.* 1968). On the other hand, during pregnancy there is also a high level of naturally occurring cell loss. This includes the placenta (as may be derived from syncytial sprouting and the appearance of trophoblast cells in the maternal circulation) as well as the fetus. With respect to the latter it is likely that, as in other mammals (Oppenheim 1981), regressive processes occur even in the nervous system

Table 2. Polyamine, conjugates and metabolite concentrations (nmol/l) in 24 samples of amniotic fluid collected at between 16 and 40 weeks gestation

Compound	Median	Range
DAP*	30	ND– 130
acDAP	240	ND– 590
Pu	10	ND– 230
acPu	280	100– 700
Cad	10	ND– 150
acCad	140	ND–1540
Isoputr+ Isoga†	40	ND– 450
Putr	20	ND– 110
Sd	30	ND– 110
N ¹ acSd	40	ND– 110
N ⁸ acSd	30	ND– 120
Sp	50	ND– 460
N ¹ acSp	50	20– 160
N ¹ ,N ¹² diacSp	550	190–1220
Percentage of total‡		
acDAP	88	62– 100
acPu	94	56– 100
acCad	95	47– 100
N ¹ acSd+N ⁸ acSd	70	37– 91
N ¹ acSp	9	2– 21
N ¹ ,N ¹² acSp	84	36– 95
Molar ratio		
N ¹ acSd/N ⁸ acSd	1.0	0.7–4.0

* $n = 30$.

† Sum of Isoputr and Isoga.

‡ Mol% of the sum of the respective free and acetylated polyamine.

ND, Not detectable.

See text for list of abbreviations.

(Precht 1984). Polyamine-degrading activity seems increased, as suggested by the high placental and maternal plasma amine oxidase levels. At present it seems impossible to discriminate between these completely opposite effects which may both lead to extracellular acPu increases.

As patients with advanced high-grade non-Hodgkin's lymphoma excrete spermine mainly as N¹,N¹²diacSp before and during successful chemotherapeutic treatment (van den Berg *et al.* 1986a), the appearance of N¹,N¹²diacSp in amniotic fluid and urine during pregnancy is probably due to cell loss. N¹,N¹²diacSp is a substrate for (FAD-dependent) polyamine oxidase from rat liver (Seiler *et al.* 1981) and human pregnancy serum (Morgan 1987), that converts it into N¹acSd. The presence of N¹,N¹²diacSp in urine may be caused by a high activity of

cytosolic acetyl transferase, or overloading of the activity of maternal serum polyamine oxidase, whereas its presence in amniotic fluid may be caused by a low activity of fetal polyamine oxidase. An increase of extracellular N¹,N¹²diacSp does not occur in all circumstances characterized by highly augmented release of polyamines, as patients with advanced acute myeloblastic leukaemia excrete undetectable amounts during successful chemotherapeutic treatment (unpublished observations). Excretion of N¹,N¹²diacSp may therefore be cell specific, i.e. depending on the intracellular ratio between N-acetyltransferase and (FAD-dependent) amine oxidases. In pregnancy it may be related to the degree of differentiation during fetal development, as from 32 weeks gestation the N¹,N¹²diacSp levels start to decrease in urine. Newborns excrete considerable amounts of N¹,N¹²diacSp up to 4 months of age. As no gestational-age-dependent alterations in the percentage of acetylation are found in amniotic fluid or in urine, the appearance of N¹,N¹²diacSp may rather be due to relatively low levels of FAD-dependent polyamine oxidase activity in the fetus and subsequently an overloading of maternal polyamine oxidase, than to relatively high activity of N-acetyltransferase. The formation of N¹,N¹²diacSp may under these circumstances be a normal detoxicative process for free Sp, which is a highly toxic compound (Rosenthal *et al.* 1952). This is confirmed by the observation that treatment of mice with polyamine oxidase inhibitors for a period of 6 weeks produced neither toxic effects nor changes in body weight or behaviour (Bolkenius *et al.* 1985).

In conclusion, to date only fragmentary knowledge of the different excretory forms of polyamines in health and disease is available. Determination of acetylated polyamines may provide more precise markers of alterations in cell proliferation, differentiation and cell loss, than do the free compounds or the acid-hydrolysed polyamines. The appearance of extracellular N¹,N¹²diacSp supports the hypothesis that unique conjugates may be produced in specific diseases (non-Hodgkin's lymphoma) and under altered physiological circumstances (pregnancy). The production of N¹,N¹²diacSp may be dependent on the degree of differentiation, coherent with maturation of the FAD-dependent polyamine oxidase activity. It is tempting to speculate that alterations in the growth and

development of the fetus will show deviant polyamine concentration profiles.

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Assessment of trace metal intake and status in a Gujarati pregnant Asian population and their influence on the outcome of pregnancy

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Summary. Dietary assessments and biochemical indices including plasma zinc and copper were determined in pregnant vegetarian and non-vegetarian Gujarati women in India at 28 weeks gestation, and the results were contrasted with those of a comparable group of Gujarati pregnant women living in Harrow. Even though the dietary intake of energy, protein and zinc was significantly lower in the Indian vegetarian Gujarati group when compared to the equivalent group in Harrow, the birthweights of the babies delivered at term were similar. The intake of zinc in the diet of the Gujarati Indian vegetarian and non-vegetarian groups was only one quarter of the US recommended intake during pregnancy and was approximately one half in the Harrow Indian groups. Even so, the plasma concentrations of zinc were similar in all dietary groups in either India or Harrow. The albumin content in the plasma of both the Gujarati Indian vegetarian and non-vegetarian was significantly reduced when compared to the equivalent Harrow dietary groups. The plasma concentrations of both calcium and sodium were significantly correlated with the albumin concentration but not with the plasma content of zinc. Copper levels were elevated to the normal range in both dietary groups of the Gujarati and were similar to the concentrations found in the Harrow groups.

The zinc status of a vegetarian population may be low because of an inadequate intake or a reduction in the bioavailability of zinc in the diet (Freeland-Graves *et al.* 1980a). During pregnancy there is an increased demand for zinc by the fetus and the low zinc status in vegetarians

could be detrimental to the developing fetus. Maternal zinc deprivation during pregnancy has been shown to cause retarded intrauterine growth in experimental animals (Hurley 1981). The lower birthweight of infants born to Asian mothers compared to whites in Britain reported

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previously by ourselves (McFadyen *et al.* 1984) and others (Grundy *et al.* 1978; Miere & Farrant 1981) may be attributable to an inadequate diet (Viegas *et al.* 1982) with a decreased bio-availability of zinc (Campbell-Brown *et al.* 1982).

Recent assessments of the dietary intake of three groups of pregnant women, i.e. vegetarian and non-vegetarian Hindu Asians and Europeans presenting at Northwick Park Hospital, showed a lower zinc content in the diets of Asian vegetarians (Abraham *et al.* 1985; Campbell-Brown *et al.* 1985). However, the mean birthweights (crude or adjusted) were similar in both Asian groups although the Muslims had somewhat heavier babies. Furthermore, there was no association between the birthweight and any of the markers of zinc status in the three groups (Campbell-Brown *et al.* 1985).

To identify whether the diet and biochemical indices had been influenced since immigration to the UK, dietary information was collected from a comparable group of Asian vegetarians and non-vegetarians of approximately 28 weeks gestation living in Gujarat, India. These results were then compared with the data of our previous study (Campbell-Brown *et al.* 1985).

Patients and methods

Healthy pregnant vegetarian and non-vegetarian Gujarati Indian women ($n = 73$) were recruited from two outpatient clinics in Baroda, Gujarat, India (SSG Hospital and Kalpana Clinic), and information collected on their dietary, social and ethnic origins in a comparable manner to our previous study. The Indian dietitian (R.A.) travelled to Gujarat where she obtained the dietary, obstetric data and blood samples. Dietary assessment was by 7-day dietary recall, and patients were divided according to their diet into vegetarians ($n = 53$) and non-vegetarians ($n = 20$). Nutrient intake was determined using food composition data (Paul & Southgate 1978). Gestational age was determined from the patient's menstrual data and by fundal height measurement. Social class was based on husband's occupation using the Registrar General Classification. Information on religion, parity, weight, height, age, complications during pregnancy, haemoglobin, previous obstetric history, worm infestation and tobacco smoking and chewing was also obtained. Details of the delivery and birthweight

of babies were subsequently collected by the doctor (M.P.) from 31 subjects. These follow-up data are limited because some women moved away and others were difficult to trace because of the large throughput in the Obstetric Department of the Hospital. Non-fasting lithium-heparin blood specimens were collected from 62 patients during their routine visit to the outpatient department, centrifuged at 800 *g* for 15 min and the plasma frozen at -4°C before transport to Harrow for analysis of zinc, copper (Scott *et al.* 1983) and other biochemical indices. Comparison of these results was made with those of the vegetarian and non-vegetarian Harrow Asian population who were of approximately 28 weeks gestation (Campbell-Brown *et al.* 1985) and most of whom were Hindus of Gujarati descent who had emigrated to Britain during the previous decade.

Results

Maternal and infant characteristics

The Indian women in Gujarat were about 10 kg lighter than the Harrow Indians (Table 1). The mean birthweights in both the vegetarian and non-vegetarian Gujarati groups were similar (Table 1). If adjustment is made for the maternal size of the Gujarati Indian mothers, the birthweight of these Indian babies is relatively greater than that of those born in Harrow.

Approximately half of the Gujarati Indians were of social class IV or V compared with 12% of the Harrow Indians. Approximately 50% of the Harrow Indians were of social class I or II compared to only 10% of the Gujarati Indians.

Dietary data

The mean daily dietary intakes of the two groups of pregnant Asian women living in Gujarat and Harrow are shown in Table 2. The energy and protein intakes of both vegetarian and non-vegetarian Gujarati Indian women were lower than those of similar groups of pregnant Harrow Indian women. The proportion of total calories derived from protein sources is the same among the Gujarati Indian women seen in Gujarat and Harrow. The diet of the Gujarati Indian women is more 'prudent' with a lower fat (36% cal) content and higher carbohydrate (60% cal) and fibre content (14.4 g/1000 kcal).

Table 1. Maternal height and weight in the two Hindu Asian populations in Gujarat and Harrow at 28 and 20 weeks gestation respectively and infant birthweight

	Gujarat		Harrow	
	Vegetarian	Non-vegetarian	Vegetarian	Non-vegetarian
	(n=49)	(n=13)	(n=59)	(n=33)
Height (cm)	150 (5.1)	149 (6.7)	155 (5.8)	156 (4.8)
Maternal weight (kg)	46.1 (7.0)*	46.6 (5.0)*	55.1 (9.3)*	56.5 (7.6)*
	(n=24)	(n=7)	(n=59)	(n=33)
Maternal weight (kg)	46.9 (8.2)	46.2 (4.6)	55.1 (9.3)	56.6 (7.6)
Birthweight (g)	2885 (547)	2904 (383)	2905 (517)	2926 (635)

Results are means (SD). Statistical significances are calculated between places within similar dietary groups. * $P < 0.001$.

Table 2. Mean daily dietary intake of pregnant vegetarian and non-vegetarian Gujarati Indians compared to the corresponding dietary groups in Harrow Indian pregnant population

	Gujarat		Harrow	
	Vegetarian (n=53)	Non-vegetarian (n=20)	Vegetarian (n=59)	Non-vegetarian (n=33)
Energy (kcal)	1320 (529)*	1460 (635)*	1970 (523)*	2165 (460)*
Protein (g)	38 (17)*	43 (20)*	57 (16)*	75 (19)*
Zinc (mg)	5.0 (2.2)**	5.7 (2.5)*	7.5 (2.1)*	10.2 (3.0)*
Copper (mg)	1.2 (0.6)**	1.3 (0.6)***	1.5 (0.7)**	1.8 (0.8)***
Iron (mg)	9.7 (5.5)	11.4 (5.9)	11.1 (4.6)	12.8 (4.5)
Fibre (g)	18.6 (9.2)***	21.9 (11)	23.1 (6.9)***	22.8 (6.8)

Results are means (SD). Statistical significances are calculated between places within similar dietary groups. * $P < 0.001$, ** $P < 0.02$, *** $P < 0.05$.

Zinc intake among the Gujarati Indians is only 25% of the 20 mg recommended during pregnancy in the USA, while intakes of iron and copper are comparable to those of Harrow Indians. The phytic acid:zinc ratios are similar in the Gujarati Indian vegetarian and non-vegetarian diet (15.9 and 14.0 respectively) confirming (Abraham 1983) that non-vegetarianism merely implies the addition of small amounts of meat or fish to the diet. Thus these non-vegetarian diets cannot be compared to a European non-vegetarian diet.

Plasma zinc and copper concentrations

The plasma zinc concentrations were similar in both the vegetarian and non-vegetarian Asian women, whether living in Gujarat or Harrow (Table 3) and were reduced below the normal range (10.5–18.0 $\mu\text{mol/l}$). The mean copper levels in the plasma were increased above the

normal range (12.6–18.5 $\mu\text{mol/l}$) and were similar in all four groups although the range of values in the Gujarati women was wide. In the 31 subjects from whom birthweights of babies were obtained, the mean concentrations of both zinc and copper were similar to the total group (Table 3); in addition, statistical analyses show that there is no evidence of a real difference between the means.

Biochemical indices

The biochemical indices determined in the plasma samples of the vegetarian and non-vegetarian Asian pregnant women living in either Gujarat or Harrow are shown in Table 4. The mean albumin concentration was significantly lower in the plasma samples of both Gujarati groups when compared to their corresponding dietary group living in Harrow. There were significant correlations between the albumin con-

Table 3. Plasma concentrations of zinc and copper at approximately 28 weeks gestation in Indian women

	Gujarat		Harrow	
	Non-vegetarian	Vegetarian	Non-vegetarian	Vegetarian
All women				
Zinc ($\mu\text{mol/l}$)	8.3 (1.6) (<i>n</i> =13)	8.6 (1.6) (<i>n</i> =49)	7.9 (1.3) (<i>n</i> =23)	8.4 (1.0) (<i>n</i> =41)
Copper ($\mu\text{mol/l}$)	30.9 (10.5) (<i>n</i> =13)	30.9 (12.0) (<i>n</i> =49)	29.4 (5.5) (<i>n</i> =23)	30.4 (4.1) (<i>n</i> =41)
Women with known infant birthweight				
Zinc ($\mu\text{mol/l}$)	8.7 (1.7) (<i>n</i> =7)	8.3 (1.7) (<i>n</i> =24)		
Copper ($\mu\text{mol/l}$)	28.4 (9.7) (<i>n</i> =7)	29.4 (10.6) (<i>n</i> =24)		

Results are mean (SD) values.

Table 4. Serum biochemical indices in Gujarati and Harrow Indian vegetarian and non-vegetarian pregnant patients at 28 weeks gestation

	Gujarat		Harrow	
	Non-vegetarian (<i>n</i> =13)	Vegetarian (<i>n</i> =49)	Non-vegetarian (<i>n</i> =23)	Vegetarian (<i>n</i> =41)
Alkaline phosphatase	130 (90)	107 (39)	105.7 (30)	109.5 (41)
Calcium	2.12 (0.11)	2.10 (0.21)	2.20 (0.11)	2.19 (0.11)
Total protein	63 (5.1)	61 (7.7)	61 (4.4)	62 (4.6)
Albumin	31.9 (3.0)*	30.8 (3.9)**	34.9 (2.2)*	35.4 (2.3)**
Phosphate	1.11 (0.16)	1.20 (0.19)	1.06 (0.21)	1.03 (0.25)
Bilirubin	5.6 (1.4)	6.3 (4.8)	6.4 (3.2)	6.8 (3.8)
Sodium	133 (4.1)	132 (7.6)	138 (6.1)	137 (2.5)
Potassium	3.8 (1.1)	3.7 (0.9)	4.03 (0.5)	4.19 (0.4)
Urea	2.47 (0.74)	2.6 (0.81)	2.9 (0.7)	2.7 (0.6)

Results are mean (SD) values. Significances are calculated between places within similar dietary groups.

* $P<0.002$, ** $P<0.001$.

centrations and the concentrations of sodium and calcium in both groups ($r = 0.635$, $P<0.01$ and $r = 0.802$, $P<0.01$, respectively). However, there were no significant correlations between the concentrations of albumin and zinc in the plasma of these Gujarati Indians. There were similar mean activities for alkaline phosphatase in each of the four groups. Sample cumulative distributions on a probability scale were drawn of (a) zinc, (b) sodium, (c) calcium, (d) albumin, and (e) alkaline phosphatase for the four groups. These indicated non-normality in the distributions of their values (Fig. 1) but were fitted adequately by skewed, unimodal theoretical frequency curves (Johnson 1949). Thus although non-normal, there is no evidence for more than one population of values within a given group.

Discussion

The dietary intake of zinc in both the vegetarian and non-vegetarian Gujarati Indian groups was lower than the corresponding dietary groups in the Harrow Indian women. Despite this low daily intake of zinc (5.0 (SD 2.2) mg and 5.7 (SD 2.5) mg respectively), the birthweights of the babies born to the Gujarat women were similar to those of the Harrow Hindu women whose dietary zinc intake was almost double (i.e. 10 mg). If the quality of pregnancy is assessed solely on the birthweight of babies we would conclude from this study of the Gujarati Indian that there was an adequate nutritional intake in these subjects despite the low dietary intake of zinc and other nutrients. Indeed after adjust-

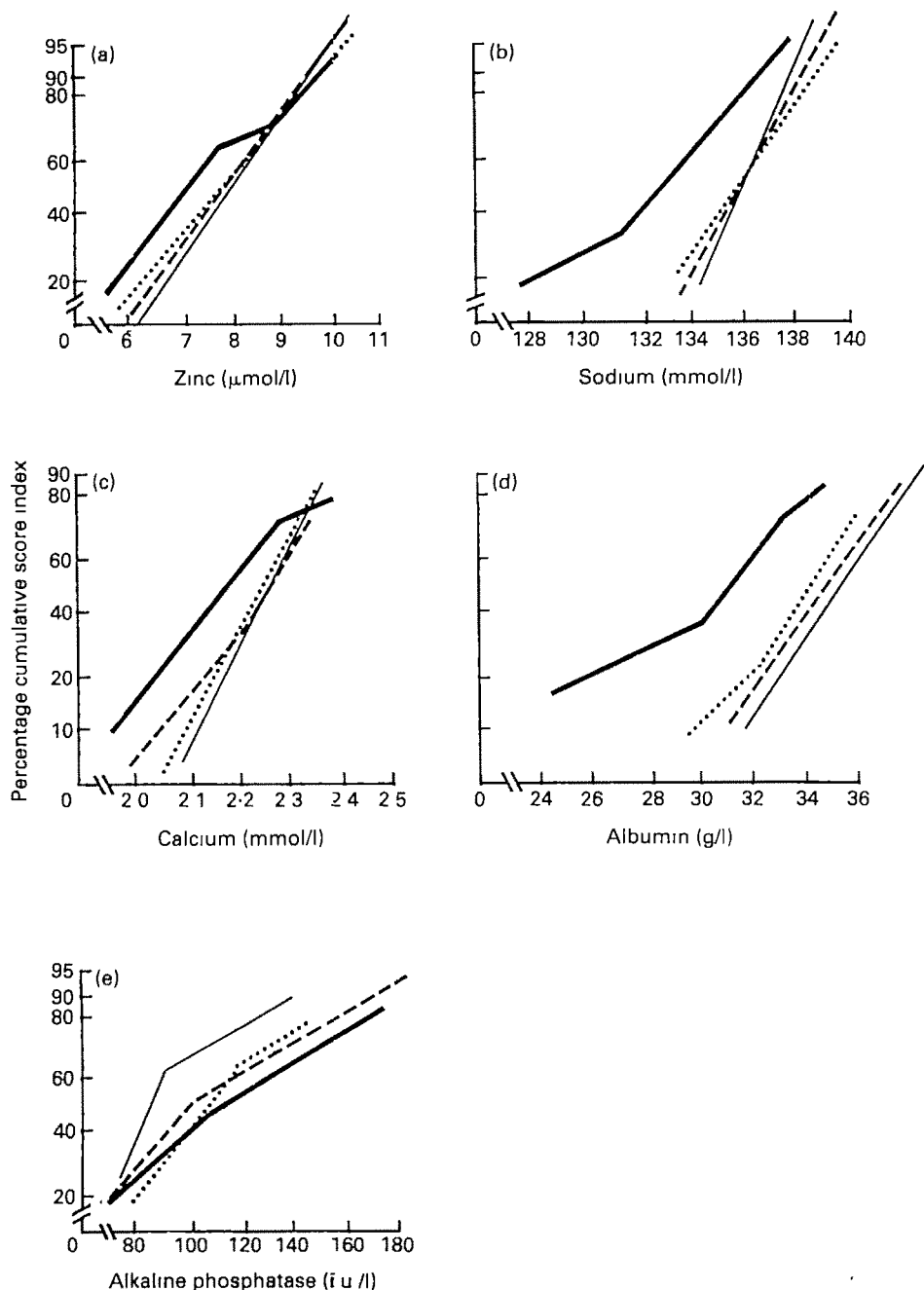


Fig. 1. Sample cumulative distribution curves for (a) zinc, (b) sodium, (c) calcium, (d) albumin, and (e) alkaline phosphatase for Hindu vegetarians, i.e. Gujarati and Harrow vegetarians, and non-vegetarians, i.e. Hindu and European meat eaters. —, Gujarati Hindu vegetarians; —, Harrow Hindu vegetarians; ..., Harrow Hindu meat eaters; —, European meat eaters.

ment of birthweight for the lower maternal weights in the Gujarat it appears that these babies were no lighter. Also preterm delivery would be expected to be more common in India

so this does not bias birthweight data in favour of Gujarati. The availability of zinc for intestinal absorption is believed to be 10–40% (WHO 1973) and it has been shown to be better from

animal foods than vegetable foods (Swanson *et al.* 1983), but there may well be an adaptation in the vegetarian by increased absorption (Freeland-Graves *et al.* 1980b; Kies *et al.* 1981; Anderson *et al.* 1981). Most assessments of vegetarians' diets, whether pregnant (Hunt *et al.* 1978; King *et al.* 1981; Tuttle *et al.* 1985), or not (Freeland-Graves *et al.* 1980a; King *et al.* 1981; Anderson *et al.* 1981) have reported significantly reduced intakes of zinc in their diets. However, in these studies no information has been given of the outcome of the pregnancy with reference to the babies' birthweights. Only one other study (Mittal *et al.* 1982) has assessed the intake of zinc in Gujarati Indians during the second trimester of pregnancy and reported an intake of 9.9 mg. These intakes which are considerably less than the US recommended daily allowance suggest that either almost all pregnant women are zinc deficient or that the recommendation is excessive—the latter is probably the case.

The zinc contents of either leucocytes or plasma are the two biological materials reported to be pertinent markers of the zinc status of an individual. Zinc is an important component of many intracellular enzymes, so that its measurement in leucocytes may reflect cellular levels. By contrast the zinc in plasma attached to transport proteins is labile, such that the total plasma content may be modified by a number of factors including diurnal variation (Scott *et al.* 1983) and albumin concentration (Stanwell-Smith *et al.* 1983). But providing careful control is made for these biological alterations by collecting blood specimens when diurnal differences will be minimized (i.e. between 11.00 and 14.00 hours), and determining plasma proteins, plasma zinc will reflect the balance between cells and therefore whole body zinc status. The lower values for zinc in the plasma during pregnancy which decrease with gestational age have been related to hypoalbuminaemia, although positive correlations have been calculated in some studies (Campbell-Brown *et al.* 1985) but not in others (Tuttle *et al.* 1985). No differences were detectable in plasma concentrations of zinc at 28 weeks between any of the dietary groups whether living in India or Harrow despite the marked hypoalbuminaemia which occurred in all patients living in India. This would indicate that the zinc status is similar in the Gujarati and Harrow vegetarian and non-vegetarian at 28 weeks. Alterations in the affinity of plasma proteins for zinc may occur during pregnancy (Kasarskis & Schuna 1980)

with diminished albumin binding of zinc (Giroux *et al.* 1976). It is unclear why the values for zinc in plasma were not proportionally lower in the dietary groups living in India because of this hypoalbuminaemia. Similar mean values for plasma zinc in both Harrow and Gujarati women were noteworthy. Supplementation with iron during pregnancy is widespread, and could possibly cause antagonism in the absorption of other trace metals such as zinc. Since iron supplementation was given only to the Harrow Indian groups, this may have lowered the circulating concentration of zinc in these subjects. Previous studies have indicated a decrease (Meadows *et al.* 1983) or no change (Shelton *et al.* 1985) in circulating zinc concentration of patients who had been supplemented with iron. Assessment of the data using sample cumulative distribution analysis did suggest there might be two different populations present, but this was not confirmed with further statistical analysis.

The increase in plasma copper concentrations during pregnancy has been well reported (Tuttle *et al.* 1985) and is caused by a parallel increase in its major binding protein caeruloplasmin. Although information on the birthweights was only collected for approximately 50% of the enrolled patients, there was no obvious relation between the plasma copper concentration at 28 weeks and birthweight.

Our results indicate that a diet depleted of zinc by approximately three-quarters of the US recommended dietary allowance during pregnancy was not detrimental to the outcome of pregnancy if birthweight is the criterion of a successful outcome. Supplementation of the Indian Gujarati population with zinc during their pregnancy was clearly not indicated as there was no impairment of fetal development in this population when compared with the birthweights of either the Harrow Indian or white population.

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Did Princess Charlotte die of pulmonary embolism?

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Summary. The cause of death of the Princess Charlotte of Wales in 1817 has been attributed to postpartum haemorrhage. Although an autopsy was performed, pulmonary embolism was not diagnosed because this condition was not recognized until 1846, but the clinical description of her last hours clearly points to pulmonary embolization as the most likely cause of death.

Princess Charlotte, heiress to the throne of England, daughter of George IV and wife of Prince Leopold, died in childbirth in November 1817. Richard Croft, her obstetrician, committed suicide 3 months later. Since her child was stillborn, her death augured the end of the House of Hanover, led to the birth of Victoria and so the Victorian era in England. Her widower became King of Belgium and it was his grandnephew who resisted the advance of the German army through the low countries in 1914, so delaying Ludendorff's plans significantly.

Although nearly 170 years have passed, the debate continues about the cause of her death. Fox (1917) suggested that the cause of death was pulmonary embolism. This explanation was rejected because at autopsy her lungs had been described as normal. Holland (1951) attributed Charlotte's death to postpartum haemorrhage. This theory was reiterated and supported more recently by Dewhurst (1980). This paper presents circumstantial clinical and historical data that the Princess Charlotte died of pulmonary embolization and that the supposition by Fox (1917) was, in fact, correct.

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Materials, methods and sources

Information about the pregnancy and labour of the Princess Charlotte was available immediately after her death both from official reports and from press reports. In 1951, family descendants of the obstetrician concerned in the case, Sir Richard Croft, made available to Sir Eardly Holland new material discovered in an attic in the family home. It was published by Crainz (1977). Many of the details of the pregnancy and labour may, therefore, be pieced together from this evidence, and we have found it useful to summarize this account.

Pregnancy of the Princess Charlotte

Princess Charlotte married Leopold of Saxe-Coburg on 2 May 1816. The marriage was happy and contented and for the first time in a tumultuous life Charlotte was fulfilled. The couple moved from London to Claremont in Surrey. Following a miscarriage in July 1816, Charlotte became pregnant again in early 1817. Her last menstrual period was on 9 January. Sir Richard Croft, one of the leading accoucheurs in London, was appointed as the Royal Obstetrician. Her physician was Matthew Baillie and the consultant appointed was Dr John Sims.

Records of Charlotte's pregnancy and labour are well documented by nine of the original letters from Charlotte to Croft and Croft's labour notes (Crainz 1977). Charlotte adhered to the instructions given to her by Dr Croft and Dr Matthew Baillie, both of whom had based their

practice of midwifery on that of Thomas Denman, Croft's father-in-law and the most eminent and influential British obstetrician of the day. Charlotte's prescribed diet was low in iron and other minerals. She had periodic phlebotomies, but each phlebotomy was associated with 50–150 ml of blood loss. However, it is likely that she ate normally as would healthy, young women of her class so that the assumption that the prescribed diet and the bleedings led to anaemia may not be warranted.

Sir Richard Croft moved into residence at Claremont to be in readiness for the impending labour in mid October 1817 when Charlotte was 39 weeks pregnant. Labour did not begin until 19.00 hours on 3 November when she was 42 weeks and 2 days from her last period. There were irregular and weak contractions and soon after the membranes ruptured. To illustrate the course of the labour, the available information has been incorporated into graphic form as described by Friedman (1981) (Fig. 1). Four hours after the membranes ruptured, Charlotte was examined for the first time by Croft who found the os uteri to be the size of a halfpenny, i.e. 2.5 cm. In the early morning of Tuesday, 8 h into labour, Charlotte had a violent attack of retching. Croft, thinking that this meant her labour was advanced, summoned the six officers of State as well as his previously agreed consultant, Matthew Baillie, to the Claremont residence. However, labour continued to progress slowly. Charlotte was kept fasting throughout the entire labour. She was examined again 16 h after the commencement of labour when the os

was the size of a crown piece (approximately 3.5 cm) with thin margins. Croft, finding the uterus acting irregularly and anticipating the possibility of requiring instruments if labour continued to progress slowly over the next 12 to 18 h, became concerned. He wrote a letter to Dr John Sims, the senior consultant, but held back from sending it when Charlotte's labour began to improve. At 18.00 hours, 23 h after labour had started, Croft made his third examination and found the cervix was nearly fully dilated and at 21.00 hours the cervix was fully dilated and for the first time an ear of the child could be felt. The duration of the first stage of labour was 26 h.

At this point, Charlotte was in good spirits. Croft, at the completion of the first stage, may have been a little anxious, because he now sent the previously written letter to Sims and requested his immediate attendance. The uterus continued to contract irregularly and weakly. Sims arrived at 02.00 hours on 5 November, 5 h after the beginning of the second stage. He met privately with Dr Baillie and Dr Croft and was informed of the conduct of Charlotte's labour. Croft proposed that Sims be introduced to Charlotte, but Sims and Baillie felt that this might unnecessarily upset the Princess and she was, therefore, not told of his arrival. Instead, Sims remained in the adjoining room where he was regularly told of the progress of the labour and was at hand to see Charlotte whenever it might be thought necessary. Instruments were in readiness but, as Croft wrote, 'the employment of them never became a question'.

At midday on 5 November, 15 h after the com-

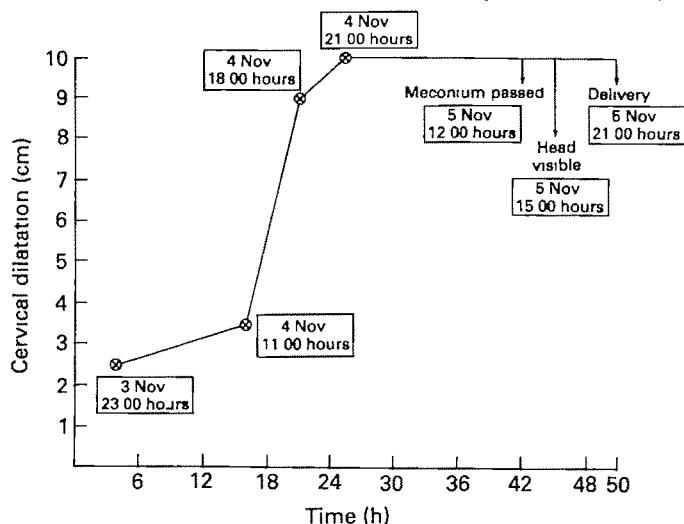


Fig. 1. A Friedman curve of Princess Charlotte's labour.

mencement of the second stage, a dark green discharge appeared, and all three physicians in attendance suspected that the child might be dead. After 18 h of second stage the child's head began to press upon the external parts, and after 24 h of the second stage, a 9 lb male child was born 'without a mark of life' by the action of Charlotte's pains alone. The umbilical cord was very small and dark green. Sims attempted to resuscitate the child by every known method. Brandy was put into the child's mouth and its lungs were inflated before the cord was cut. The child was then immersed in a warm bath and rubbed with salts and mustard, but 'no animation' was ever restored. The labour had lasted for 50 h; 26 h in the first stage and 24 h in the second stage with 6 h of perineal pressure. Labour in the second stage was as inefficient as it had been in the first. Charlotte had shown no signs of deficient strength during the entire labour. She was frequently up and walking about the room during the daytime and remained up until a short time before delivery.

The third stage of labour now began. The uterus continued to act irregularly, and 10 min after the birth of the child, Croft suspected an hour-glass contraction of the uterus with the placenta retained in its upper part. The placenta could not be felt and the uterus remained high. He informed Sims who was trying to resuscitate the child in the next room. Twenty minutes later a bloody uterine discharge appeared and the doctors agreed that the placenta should be removed manually. Croft met with no difficulty until he came to the contracted part of the uterus which would only allow three fingers to pass. By applying constant pressure, the contraction eventually gave way and the placenta was peeled off 'with considerable facility', brought down and left in the vagina as was the custom in that day (Dewhurst 1980). The uterus contracted down strongly and only a moderate amount of blood appeared to be lost. Immediately after this, Charlotte complained of acute abdominal pain which recurred to a lesser extent at intervals until 22.00 hours. As Charlotte complained of the placenta giving her 'great inconvenience' and as the placenta was protruding from the vagina, Croft removed it and this was followed by a very moderate discharge of 'either blood or coagulum'. By palpation through the bandages covering Charlotte's abdomen, the uterus appeared to be contracted. The duration of the third stage of labour was 1 h.

Charlotte was informed about the death of her child. She appeared surprisingly well after her long ordeal. She took nourishment. Her pulse was firm and steady, not exceeding 100 beats/min. By 23.45 hours she was not as well as she had been before. She vomited and complained of a singing noise in her head. She appeared to be irritable. Her agitation gradually increased so that between 00.30 and 00.45 hours she is reported to have been extremely restless and had great difficulty with breathing 'with spasmodic affection of the chest'. According to Matthew Baillie's notes, 'about a quarter before one, she complained of great uneasiness in her chest, and she began to breathe with great difficulty. Her pulse was very feeble and irregular, and she became extremely restless, not being able to remain a single moment in the same posture. This very alarming state continued, and kept increasing, till half past two in the morning when Her Royal Highness expired'. Her death therefore occurred a little more than 2.5 h after the onset of her respiratory difficulty.

Princess Charlotte's labour in the light of contemporary practice

The management of Charlotte's labour must be judged in the light of midwifery practice of the day. The chief criticism that has been made is that Sir Richard Croft allowed labour to go on so long without using forceps, but by the principles and rules of contemporary practice, he was not wrong. Examination of the standard textbooks reveals the astonishing degree of conservatism that prevailed at that time (Denman 1817). William Smellie had been a proponent of forceps with certain well-defined indications (Smellie 1779). After he retired in 1760, forceps began to be used far too freely, often by unskilled men, and much damage resulted. This period of overzealous use of forceps extended into the time of Sir Thomas Denman whose teachings dominated British obstetrical thought and practice in the late 18th and early 19th centuries. With Denman came a new era of ultraconservatism and non-interventionist obstetrics. In his book, Denman (1801) states his philosophy strongly:

'But if we compare the general good done with instruments, however cautiously used, with the evils arising from their unnecessary and improper use, we might doubt whether it would not have been happier for the world if no instru-

ment of any kind had ever been contrived for, or recommended in the practice of midwifery'.

Under the heading of third stage of labour, Denman (1801) wrote:

'For many years I have made it a rule to leave the placenta, whether naturally or artificially separated, to abide in the vagina for one hour after it was voided out of the cavity of the uterus'.

Concerning concealed postpartum haemorrhage, he wrote:

'Coagula are supposed to keep up a distention of the uterus and cause a haemorrhage; and it is recommended to introduce the hand to dilate the cervix and clear them out from the uterus. This is not necessary and I have never practiced it; nor even troubled myself with the state of the uterus unless it was inverted after the placenta was brought away; but have left whatever coagula it contained to be expelled by its own action'.

Had Croft followed the teaching of Smellie and not the conservative reactionism of his father-in-law, this labour may have resulted in the birth of a live child. It is clear, however, that the main incidents of Charlotte's labour were managed strictly according to Denman's teaching.

Dewhurst (1980) discusses what may have happened if another of Denman's protégés and one of Sir Richard Croft's colleagues on the Staff of the Royal Maternity Charity had been at Charlotte's confinement. This was David Daniel Davis who 3 years later delivered the mother of Queen Victoria. Davis was one of the minority of accoucheurs of the time who favoured the selective use of instruments to expedite delivery. Instruments had been used at the birth of his own child causing injury and eventual death. Davis redesigned forceps by enlarging the blades and decreasing the compression effect and published these innovations in 1825 as *Elements of Operative Midwifery* (Davis 1825). As Dewhurst (1980) says, 'It is difficult to believe that despite the enormous influence of Thomas Denman's teaching, a true leader of the profession would not have intervened earlier and possibly saved the child's life'.

Autopsy

An autopsy was performed the following day by David Drundas and Everard Home, both Sergeant Surgeons to George IV, and both Masters of the College of Surgeons. It is noteworthy that

Dr Baillie, who was the author of the definitive text on pathology at the time (Baillie 1793) did not attend the autopsy.

The main findings noted at the autopsy (Crainz 1977) were: (1) Two ounces of sanguineous pericardial fluid; (2) 1500 ml of liquid in the stomach; (3) one pound of coagulated blood in the uterus that retained its hour-glass configuration; (4) a benign teratoma of the right ovary; (5) the child appeared normal.

Discussion of the aetiology of Charlotte's death

The cause of Charlotte's death has been attributed to cardiovascular collapse following postpartum haemorrhage both by Holland and by Dewhurst. The chronic anaemic state induced by an inappropriate diet and several phlebotomies is considered to have predisposed to this collapse. The description of the labour indicates that even with a liberal interpretation of the blood loss, Charlotte lost no more than 3 pints of blood, if that much. There is no mention of severe blood loss during the third stage of labour in the contemporary descriptions and the autopsy did not show more than a 'pound of coagulum' in the uterus. Charlotte's pulse rate after the third stage was less than 100 beats/min and the volume was described as good. In late pregnancy, she was described as healthy and in good spirits. The proposition that Charlotte began labour already anaemic does not seem to be supported by the available facts. Periodic small phlebotomies, of the volumes recorded, are well compensated in healthy individuals, so that it is likely that Charlotte's normal young bone marrow would have kept her haemoglobin at satisfactory levels. To invoke an iron deficiency anaemia based on the prenatal diet prescribed by Dr Croft, is to reach beyond the bounds of reasonable probability. It seems quite unlikely therefore, that blood loss superimposed on anaemia could have been the cause of Charlotte's death.

Other causes of death have been suggested. Chorioamnionitis resulting from prolonged rupture of membranes has been suggested (Dewhurst 1980) but there is little evidence in the clinical description to support such a diagnosis. Porphyria is another possibility. Although George III is likely to have had porphyria and even Charlotte's father, George IV, may have had this disease, there is no evidence that Charlotte exhibited any sign of porphyria.

The description of Charlotte's last 2.5 h is highly suggestive of severe respiratory distress, secondary to hypoxia. The rapid onset of agitation, restlessness, and thrashing about in bed are, in modern clinical practice, urgent indications to rule out pulmonary embolization when these symptoms occur in a postpartum patient. Of the several possible aetiologies of Charlotte's hypoxia, two deserve serious consideration: pulmonary embolism and aspiration of stomach contents.

Pulmonary embolism was not recognized until Rudolph Virchow described the pathology in 1846, almost 30 years after Charlotte's death (Virchow 1856). In his first paper on the subject, he pointed out that up until that time, occlusion of the pulmonary arteries by clotted blood had been recorded in only a few cases. The reason for this was that differentiation between pre- and post-mortem clots was difficult, and also because no one attached any significance to clots found in the pulmonary arteries at autopsy. In 1817, the finding of clots would therefore not have been recorded in an autopsy report, even in the unlikely event that the arteries had been opened for deliberate inspection. Interestingly, Matthew Baillie was the author of one of the first books in which autopsy pathology was treated as a distinct subject. His *Morbid Anatomy of Some of the Most Important Parts of the Human Body*, first published in 1793, was for many years the definitive text book in its field. The second edition of 1812 was the most recent at the time of Charlotte's death. None of its many autopsy descriptions contains a word about opening the pulmonary artery.

Fox (1917), 100 years after the event, suggested that the cause of Charlotte's death was pulmonary embolism. This explanation was rejected at the time because the autopsy described the lungs as being normal. In fact, those who performed autopsies in 1817 had no knowledge of pulmonary embolism, and lungs affected by it would have looked quite normal to them. A large central thrombus in the pulmonary artery would certainly have gone undetected. An 1817 autopsy report of normal lungs does not rule out what we consider to be a most likely aetiology when a young woman, following a difficult labour lasting 50 h, rapidly develops symptoms suggestive of hypoxia and cardiac arrhythmia, and then dies within 2.5 h of the onset of those symptoms.

The second possibility is that of aspiration of

stomach contents, but this seems less likely. Most probably, the finding of 1500 ml of fluid in the stomach indicates acute gastric dilatation. Patients with such a condition often aspirate even without vomiting. Actually Charlotte vomited at least once. We may assume that the lungs were sliced at autopsy, since Baillie's text book indicates that he did this routinely. Even in his absence, Everard Home, to whom he was related by marriage (he was the brother of John Hunter's wife, Hunter's sister was the mother of Matthew Baillie) would have used his procedure. Assuming that the normal lungs described at autopsy meant normal cut pulmonary parenchyma without evidence of gastric fluid in the bronchi, aspiration seems unlikely to be the cause of Charlotte's death. Also, it is less consistent with the clinical picture than is pulmonary embolism.

To physicians of the late 20th century a negative autopsy finding implies that a full pathological examination has been performed as we would perform it today. Just as one cannot judge Croft by the standards of late 20th century practice, one must interpret the autopsy findings within the limitations of pathological knowledge current in 1817. Since pulmonary embolization was then an unrecognized entity, the reported examination of the lungs becomes irrelevant to a discussion of the cause of death. The most significant finding in Charlotte's autopsy was the relatively small amount of blood in the uterus, an amount that was unlikely to have caused death.

We contend, therefore, that the clinical description of Princess Charlotte's last hours suggests that the most likely cause of death was pulmonary embolism.

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A double-blind randomized controlled trial on the use of prophylactic antibiotics in patients undergoing elective caesarean section

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Summary. In a double-blind randomized controlled study 232 patients undergoing elective lower segment caesarean section were randomly allocated to receive a pre-operative prophylactic dose of a combination of crystalline penicillin and chloramphenicol or a placebo. The two groups were comparable in terms of patient characteristics and operation variables. The group receiving antibiotics had significantly fewer febrile and infectious morbid events and thus spent fewer days in hospital than the group receiving the placebo.

The total number of caesarean sections performed in most hospitals has increased over the last 10 years (NIH Consensus Statement 1981). This is invariably associated with an increase in the number of elective caesarean sections. In 1986, the caesarean section rate at Harare Central Hospital, Harare, Zimbabwe, was 18% and of these, 28% were performed electively.

There is little doubt that the use of pre-operative antibiotic prophylaxis has significantly reduced the frequency of pelvic and wound infection after caesarean section (Wallace & Yonekura 1983; Polk *et al.* 1982; Itskovitz *et al.* 1979). These studies have, however, concentrated on the group undergoing the operation in labour. Patients having a planned elective procedure may not have the increased risk of infection, and the role of antibiotic prophylaxis before elective caesarean section has not been assessed adequately. Studies which have included such patients (Knottenbelt 1979; Padilla *et al.* 1983; Itskovitz *et al.* 1979; Gall 1979) have been unable to show a significant effect of prophylactic antibiotics. Unfortunately the number of patients involved in all these

studies has been very small. Gibbs *et al.* (1981) suggested that antibiotic prophylaxis may encourage the development of resistant organisms and interfere with any infection screen in the newborn infant. There is also a small but definite risk of anaphylaxis with the penicillins.

In developing countries such as Zimbabwe, however, other factors which may contribute to higher infection rate include poorer nutritional state, a rapid turnover in theatre use and relatively inexperienced operators.

It was against this background that a double-blind randomized controlled trial was designed to compare the prevalence of infectious morbidity in two groups of patients; one group received a dose of antibiotic treatment before going to theatre and the other received an identical solution containing water for injection.

Patients and methods

All patients who had an elective lower segment caesarean section before the onset of labour or rupture of membranes were eligible for the study. Before going to theatre consecutive patients were randomly allocated to receive an intravenous injection of either prophylactic antibiotics (solutions A1 and A2) or a placebo (solutions B1 and B2). The randomization was performed by the sister in charge of the ante-

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Table 1. Characteristics of patients undergoing elective lower segment caesarean section

Characteristics	Treatment groups	
	Antibiotics (<i>n</i> = 115)	Placebo (<i>n</i> = 117)
Age (years)		
Mean (SD)	28.2 (4.3)	26.5 (4.9)
Range	(19–45)	(18–39)
Number over 35 years	10	7
Pre-operative maternal weight (kg)		
Mean (SD)	73.1 (14.3)	70.9 (12.8)
Range	(45–113)	(46–112)
Number over 85 kg	20	16
Parity		
Mean (SEM)	2.5 (0.1)	2.9 (0.3)
Range	(0–10)	(0–8)
Primigravida	3	3
Parity 5 and over	12	12
Number of previous caesarean sections		
0	12	20
1	33	29
2	50	49
3 or more	20	19
Gestational age (weeks)		
Mean (SD)	38.8 (1.4)	38.5 (1.5)
Pre-operative haemoglobin (g/dl)		
Mean (SD)	11.9 (1.1)	11.8 (1.3)

There were no significant differences between the two groups for any variable.

natal ward using a randomized list of treatment numbers. The solutions were prepared every morning by the pharmacy and their identity was not known to the investigator or the ward staff until the completion of the study. Solutions A1 and A2 contained 2 mega units of crystalline penicillin and 500 mg chloramphenicol respectively. Solutions B1 and B2 contained the placebo.

The following outcomes were assessed: (i) febrile episode of $\geq 38^\circ\text{C}$ on two occasions at least 4 h apart after the first postoperative 24 h; (ii) wound sepsis, graded as (a) abnormal erythema and/or induration, (b) oozing wound without frank pus, (c) pus formation; (iii) endomyometritis as diagnosed by fever, uterine tenderness and foul-smelling lochia; (iv) pelvic abscess formation; (v) bacteraemia; (vi) number of days spent in hospital because of wound or pelvic infection.

After the operation antibiotics were prescribed only if a febrile complication developed.

Results

Between November 1986 and March 1987, 232 patients were enrolled into the study. It was expected that 314 patients would need to be enrolled for a reduction in the febrile complication rate from about 30% to about 15% to be significant at the 5% level. However, a preliminary analysis at the end of the 5-month period showed a significant improvement due to antibiotics, and continuation of the study was considered to be unethical. Comparability of the two groups in terms of patient characteristics and factors related to the operation are shown in Tables 1 and 2. Randomization was found to be satisfactory.

The prevalence of infectious complications in the two groups is shown in Table 3. When a patient developed several complications only the severest was coded.

Table 2. Operation variables in the two treatment groups

Variable	Treatment groups	
	Antibiotics (<i>n</i> = 115)	Placebo (<i>n</i> = 117)
Operator status		
Senior house officer	80	89
Registrar	27	23
Senior registrar/consultant	8	5
Anaesthetic		
General	72	73
Local (spinal/epidural)	43	44
Skin incision		
Midline subumbilical	106	106
Pfannenstiel	9	11
Meconium-stained amniotic fluid	2	0
Mean estimated blood loss (ml)	503	514
Over 800 ml	13	16
Length of operation (min)		
Mean (SD)	46 (6)	49 (5)
>60 min	15	18
<30 min	19	14
Indwelling catheter	8	7
Presence of marked adhesions	8	12
Tubal ligation	14	16

There were no significant differences between the two groups for any variable.

Table 3. Maternal morbidity—infectious outcome and days in hospital

Infectious outcome	Treatment groups		Relative risk (95% confidence interval)
	Antibiotics (n=115)	Placebo (n=117)	
Patients with morbid events <i>n</i> (%)	18 (16%)	43 (37%)	2.35 (1.48–3.72)
Patients with febrile episode <i>n</i> (%)	7 (6%)	33 (28%)	4.63 (2.36–9.10)
Wound sepsis			
Induration/abnormal red	5	3	0.59 (0.15–2.38)
Oozing wound—no pus	7	9	1.26 (0.49–3.28)
Pus formation	0	3	—
Pyrexia—no cause	6	22	3.60 (1.63–7.96)
Endomyometritis	0	6	—
Mean no. of days in hospital	5.43	6.18	—
Number spending more than 8 days in hospital	3	11	3.60 (1.13–11.49)

Febrile morbidity and frequency of morbid events were much lower in patients who had received prophylactic antibiotics (6% and 16% respectively) than in patients who had received the placebo (28% and 37% respectively). These differences were statistically significant ($P<0.001$). There were no serious infections in the antibiotic treatment group. The six women who developed moderate to severe endomyometritis were in the placebo group. None of the patients developed pelvic abscess or required a laparotomy. There were significantly more patients in the placebo group who spent more than 8 days in hospital.

Discussion

The use of a single-dose antibiotic was found to be effective in preventing febrile morbidity in patients undergoing elective caesarean section compared with patients who had received the placebo. Operator skill, type of skin incision, operation time and the degree of blood loss may all contribute to postoperative maternal morbidity but these were similar in the two groups. The occurrence of febrile morbidity in 28% of the placebo group is disturbing when one considers the elective nature of the procedure. Fortunately most of the infections (100% in the antibiotic group and 75% in the placebo group) were mild and/or responded to initial therapy. If all morbid events are taken into account then 16% of the antibiotic group and 37% of the placebo group had infectious complications.

The large number of patients with pyrexia without an identifiable cause were presumed to

be infected on the basis of their response to antibiotic treatment. Practical difficulties and financial restraints prevented a full infection screen on all patients with pyrexia.

In this small series no problem with adverse drug reaction was identified. The serious infections that were encountered were in the placebo group. The few patients who did develop sepsis in the antibiotic group were treated using the same antibiotics, penicillin and chloramphenicol, and all responded to the initial therapy. The argument of cost against routine prophylaxis may also be nullified by the reduction in morbidity both in terms of infection and with regard to duration of hospital stay.

We believe that a single dose of prophylactic antibiotics administered to our population undergoing elective lower segment caesarean section is beneficial in reducing infectious morbidity. A question that probably needs addressing further now is the choice of the most cost-effective antimicrobial agent and whether a multiple dose regimen is superior to a single dose.

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Immunoscintigraphy of ovarian endometriosis. A preliminary study

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Summary. CA-125 is a membrane antigen detected in the serum of some women with gynaecological disease including endometriosis. We attempted to use a radioiodinated anti-CA-125 monoclonal antibody, OC-125, to image ovarian endometriosis using immunoscintigraphy. Two women with significant endometriosis and normal serum CA-125 levels had positive immunoscintigraphy. In one woman, all areas of endometriosis were imaged; in the other, endometriosis in one affected ovary was seen, but uptake in the other ovary was less than the extent of the disease.

OC-125 is a murine monoclonal IgG1 antibody raised against a cell line from an ovarian serous cystadenocarcinoma (Bast *et al.* 1981). Using indirect immunofluorescence of cryostat sections, it reacts with most non-mucinous ovarian carcinomas (Kabawat *et al.* 1983a). The determinant antigen, CA-125, is probably a cell membrane glycoprotein, with a molecular weight of >200 000 daltons (Davis *et al.* 1986). It is also expressed on adenocarcinoma of the endocervix, endometrium and fallopian tube (Kabawat *et al.* 1983b), and may or may not be expressed on normal ovarian surface epithelium (Kabawat *et al.* 1983b; Nouwen *et al.* 1987).

CA-125 is shed into the circulation and measured in serum by radioimmunoassay with OC-125 (Bast *et al.* 1983). High serum CA-125 levels were detected in 82% of women with ovarian carcinoma compared with 1% of apparently healthy controls (Bast *et al.* 1983). Serum

levels may also be high in women with advanced adenocarcinoma of the endocervix, endometrium and fallopian tube (Niloff *et al.* 1984). ¹³¹I-labelled OC-125 monoclonal antibody F(ab')₂ fragments, injected into most women with ovarian carcinoma, display primary tumours and metastatic sites by immunoscintigraphy (Chatal *et al.* 1987).

Endometriotic tissue also expresses CA-125 (Barbieri *et al.* 1986) and serum CA-125 levels are mildly elevated in some women with endometriosis, especially those with extensive disease (Barbieri *et al.* 1986; Giudice *et al.* 1986; Patton *et al.* 1986). We employed immunoscintigraphy to determine whether CA-125 expression *in vivo* could be demonstrated in ovaries affected by endometriosis.

Patients and methods

Two women with endometriosis and normal serum CA-125 levels were investigated with immunoscintigraphy. The first was a 45-year-old woman who presented with right iliac fossa pain and deep dyspareunia 2 years after a total abdominal hysterectomy and left salpingo-oophorectomy for endometriosis. She did not have any menopausal symptoms and follicle stimulating hormone level was not elevated (4.6 i.u./l). On examination, there was a tender right

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pelvic mass. Serum CA-125 was normal (<6 U/ml). Pelvic ultrasonography showed a bilocular cystic ovary.

The second patient was a 29-year-old woman who presented to the Oxford in-vitro fertilization (IVF) unit with 5 years primary infertility and two previous failed IVF attempts. Four years earlier minimal endometriosis was diagnosed at laparoscopy. Further IVF cycles were associated with failure of fertilization and at the most recent oocyte retrieval, via the transvaginal approach, thick 'chocolate' material was incompletely aspirated from a 4-cm diameter left ovarian cyst. Serum CA-125 was normal (13 U/ml).

Surgery was arranged for both women who also gave informed consent for immunoscintigraphy to be performed in the preceding week. Serum CA-125 levels were measured at the Supraregional Protein Reference Unit, Cardiff (Bast *et al.* 1983). Thyroid uptake was blocked with oral potassium iodide, 120 mg daily, taken for 3 weeks starting the day before immunoscintigraphy. Skin tests for hypersensitivity to mouse immunoglobulin were performed before injection of the ^{131}I -labelled OC-125 monoclonal antibody F(ab')₂ fragments supplied by CIS (UK) Ltd. Each patient received 1 mg OC-125 F(ab')₂ fragments labelled with 74 MBq (2 mCi) ^{131}I in 100 ml normal saline given intravenously over 20 min.

Planar scintigrams were obtained at 3 and 5 days after injection in both women. Anterior, posterior and lateral pelvic and abdominal views were taken with a General Electric Maxi Camera, 400T, fitted with a medium energy collimator and interfaced to a 11/34A computer with gamma 11 V 3.3 software. Each view was acquired over 5 min on a 128×128 pixel matrix using a 30% energy window centred on the 364 KeV peak of ^{131}I . The bladder was outlined using $^{99\text{m}}\text{Tc}$ -labelled DTPA and the uptake was shown to be outside the bladder.

Results

In the first patient, immunoscintigraphy showed uptake in the right pelvis in all views (Fig. 1a, b & c). At laparotomy, the ovary was 7 cm in diameter and densely adherent to the bowel; it contained two 'chocolate' cysts. Histology confirmed endometriosis.

In the second patient, there was uptake in the left pelvis and minimal uptake on the right side

in the anterior view. The posterior view showed uptake on both sides but more marked on the left (Fig. 2a & b). At laparotomy, two 2–3 cm diameter 'chocolate' cysts were removed from each ovary; there was minimal peritoneal disease. Histology confirmed endometriosis in the right ovary but only compressed glands, debris and old blood could be seen in the sections from the left endometriomata.

Discussion

Endometriosis is best diagnosed by laparoscopy, but microscopic lesions may be missed (Murphy *et al.* 1986) and staging of the disease, based upon a subjective visual assessment of the size and number of lesions, may be inaccurate (Dmowski 1987). A non-invasive diagnostic test, sufficiently sensitive to be used as a screening procedure, does not yet exist.

Studies in patients with endometriosis have shown serum CA-125 to have a diagnostic sensitivity of 14–53% (Barbieri *et al.* 1986; Patton *et al.* 1986; Pittaway & Favez 1986) which is influenced by which upper limit of normal is used for serum CA-125 (16 or 35 U/ml). A higher upper limit improves specificity but reduces sensitivity.

The correlation between the immunoscintigraphic and surgical findings demonstrates the potential accuracy of the technique. Activity was also confirmed in all the surgical specimens. The large uptake in the first patient (Fig. 1a, b & c) may reflect the severity of the adhesions around the ovary as these are associated with elevated serum CA-125 levels (Patton *et al.* 1986). In the second patient the low uptake in the right ovary on the anterior view may reflect reduced antigen expression related to the level of endometriotic activity or absent expression in some lesions.

The failure to confirm endometriosis histologically in the left ovary in patient 2 despite histological confirmation in the other ovary is not unusual. A 'chocolate' cyst may actually be an old haemorrhagic corpus luteum, or an endometrioma which has undergone pressure atrophy, which can destroy the characteristic histological appearance of endometriosis (Haney 1987). It has been suggested that serum CA-125 levels differentiate between endometriotic and other benign ovarian cysts (Pittaway *et al.* 1987).

We have demonstrated that despite the expression of CA-125 in tissue severely affected by endometriosis, serum levels were normal. In

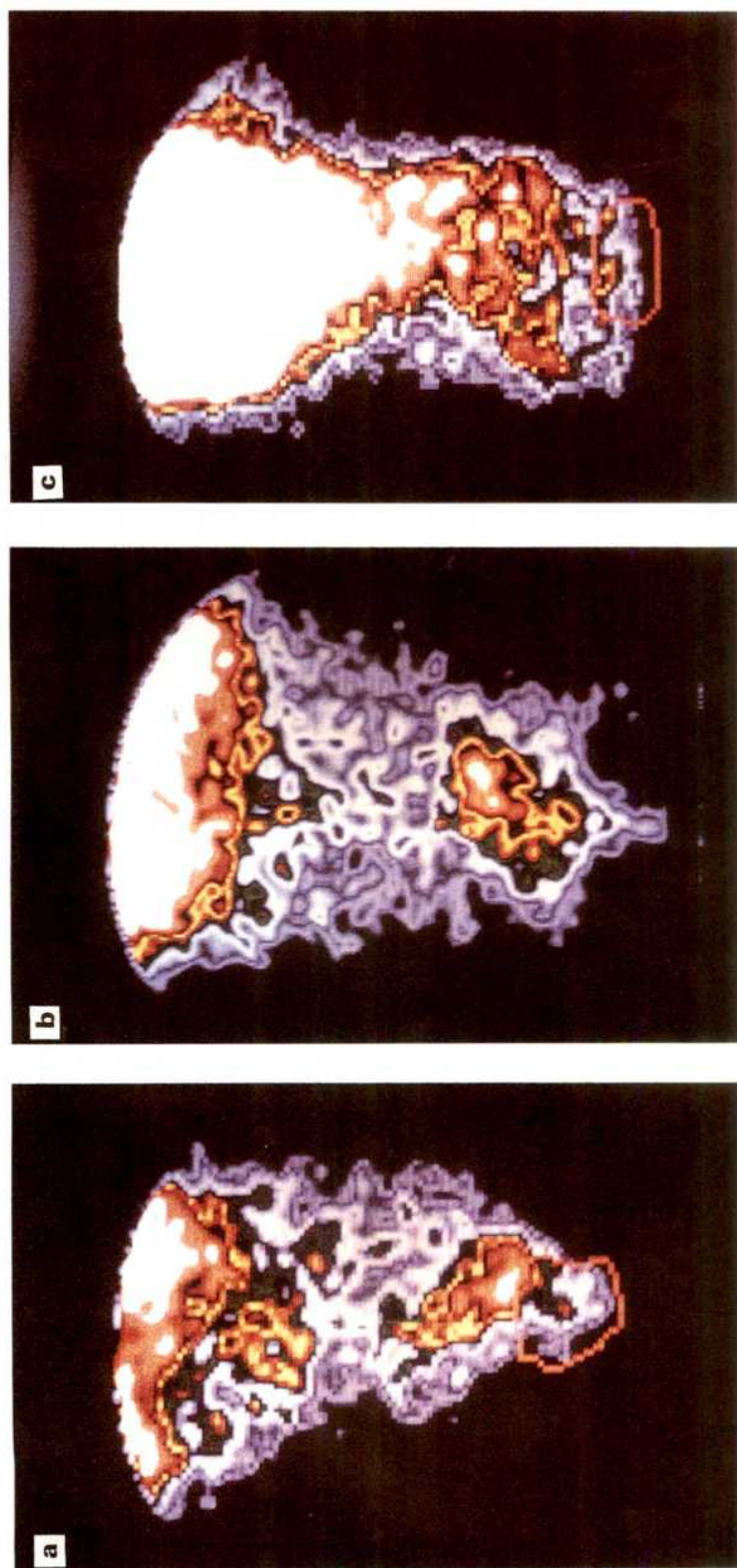


Fig. 1. Patient 1: Increased uptake on the right side of the pelvis in the anterior (a) and posterior (b) views. In the anterior (a) and lateral (c) views, the uptake is shown clearly to be outside the bladder which is outlined with technetium.

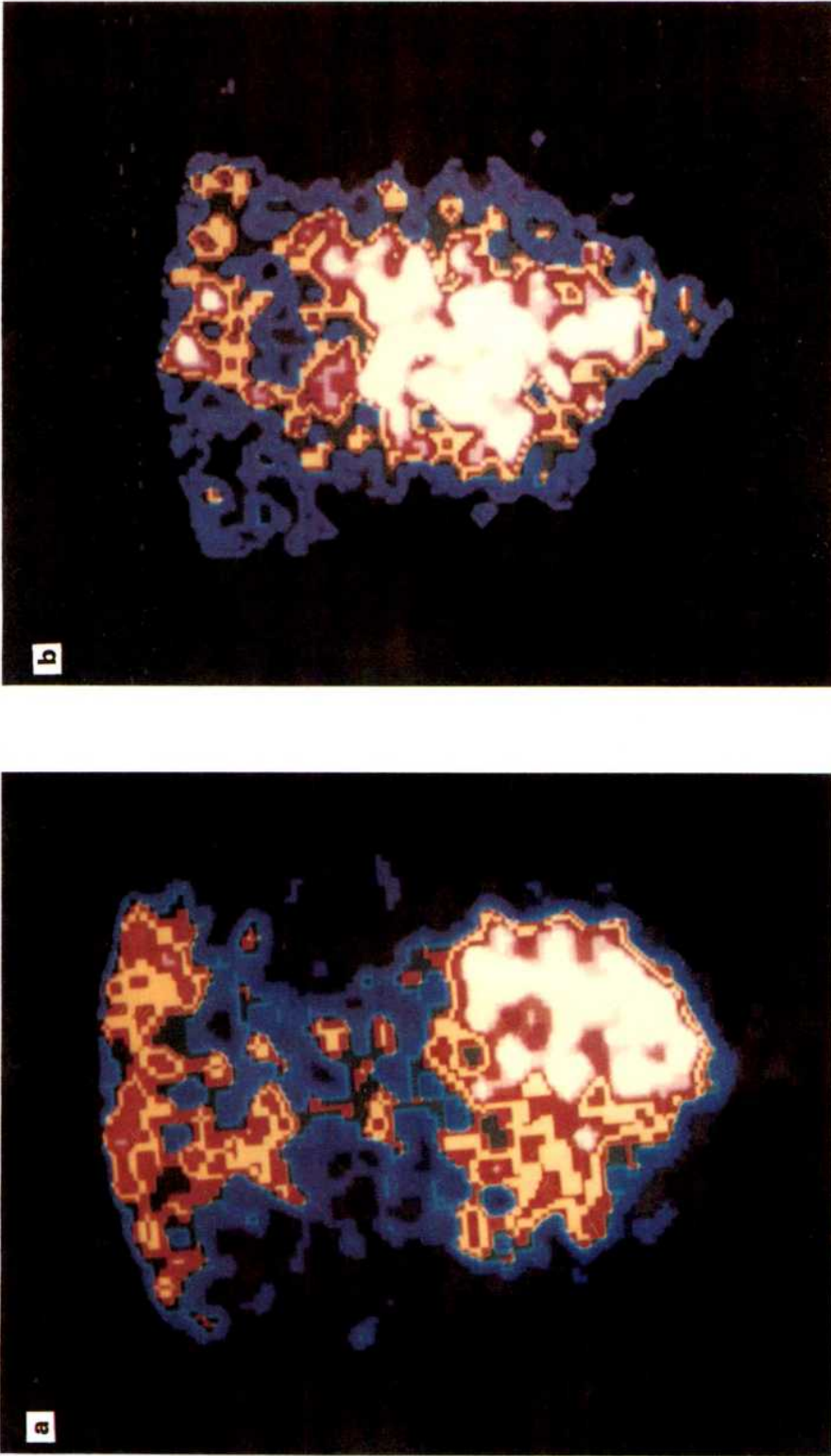


Fig. 2. Patient 2: Increased uptake in the pelvis, mainly on the left side, in the anterior (a) and posterior (b) views.

clinical practice serum CA-125 screening has proved disappointing in the diagnosis of endometriosis. Immunoscintigraphy, however, may be a more effective non-invasive diagnostic tool. It remains to be seen whether other pelvic sites can be imaged.

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The value of bladder filling in the clinical detection of urine loss and selection of patients for urodynamic testing

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Summary. Bladder filling was used to enhance the clinical detection of urine loss in 37 women who attended a general gynaecological out-patient clinic with lower urinary tract symptoms and incontinence. Videocystourethrography was performed subsequently in all women, and the clinical and fluoroscopic detection of urine loss compared. The two methods of detecting urine loss agreed in all but four women with very slight genuine stress urinary incontinence, two being detected by one but not by the other of the two tests. In only 5 of 25 patients was incontinence demonstrable clinically without bladder filling, and in only 13 of 25 was it demonstrable in the supine position. These proportions were 5 of 21 and 10 of 21, respectively, for those with genuine stress incontinence, with or without coexistent detrusor instability. Bladder filling provoked uncontrollable voiding in five women, two had detrusor instability on cystometry, and two had low peak urinary flow rates on uroflowmetry. The significance of these findings is discussed in the context of selecting patients for urodynamic testing.

From a purely practical, clinical point of view, the most important questions which need to be settled about women who complain of urinary incontinence are whether or not the incontinence warrants and is amenable to surgical correction. Most urogynaecologists believe that these questions can only be answered reliably by urodynamic testing. Although opinion is divided over what are the essential and most informative tests, many would agree, at least in principle, that all women should have urodynamic

investigations before a proposed continence operation. This recommendation stems from the belief that clinically detrusor instability (DI) can mimic genuine stress urinary incontinence (GSUI), so that without urodynamic testing inappropriate operations will be performed on women with DI, which will not only fail to relieve the incontinence, but which might make the DI worse. Therefore, some type of cystometry has been and remains the cornerstone of any urodynamic assessment, and a patient's suitability for surgery has been based as much on the absence of DI as on the presence of GSUI (Bates *et al.* 1979).

Many of these traditional views have been called into question by, amongst other things, evidence which suggests that GSUI may be aetiologically linked with DI (McGuire *et al.* 1976, 1980; Chalfin & Bradley 1982; Awad & McGinnis 1983), and that an operation which

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relieves GSUI frequently cures any coexistent DI (McGuire *et al.* 1976, 1980; Blavais & Salinas 1985; McGuire & Savastano 1985; Minini *et al.* 1985; Langer *et al.* 1986). In other words, the pre-operative identification of DI is neither a contraindication to surgery nor is it likely to alter the ultimate management of patients with clinically significant GSUI. Therefore, so far as management is concerned, it is more relevant to know whether or not an incontinent woman has GSUI which merits surgery, than to know whether or not the bladder is stable.

This shift in emphasis is of more than academic importance. District general hospitals frequently lack facilities for urodynamic investigations so that most women have to be referred to specialized centres where the waiting time for a test is usually several months. If, however, the presence of GSUI could be established reliably by clinical means in an outpatient clinic, a considerable proportion of women referred to gynaecologists with stress/urge symptoms could be satisfactorily assessed without recourse to costly testing. This study compares clinical assessment (aided if necessary by simple bladder filling) with videocystourethrography (VCU).

Patients and methods

All patients referred with lower urinary tract symptoms and incontinence to the gynaecology clinic at Greenwich District Hospital between December 1986 and September 1987 were included in this study. Patients referred to the gynaecology clinic at Lewisham Hospital were included thereafter until all patients from Greenwich had undergone urodynamic investigation, and then the study was closed. All patients were seen and examined by the author, and there were no exclusions.

After the history was taken and a general gynaecological examination carried out, patients were examined in the supine and erect positions during coughing and straining. If urine loss was not detected, the bladder was emptied, the urine volume measured, and the bladder filled with normal saline from a 500-ml sterile infusion bag using an intravenous giving set and a 12F red-rubber catheter. The 'tap' on the giving set was fully opened and the bag held at shoulder height about 60 cm above the patient. The volume infused and any urgency during filling were noted, but the infusion rate was not measured. Filling was continued until the

patient felt that the bladder was full, whereafter she was examined, as before, in the erect and supine positions. A clinical diagnosis of GSUI was made if water was lost coincidentally with coughing/straining; DI was diagnosed clinically whenever a patient voided uncontrollably during the examination. A catheter specimen of urine was sent for culture if urine analysis showed protein or if the patient complained of symptoms of frequency or urgency.

Each patient was referred to King's College Hospital for routine urodynamic investigations which consisted of VCU as described by Bates & Corney (1971) and, in some cases, urethral pressure profilometry as described by Hulton & Stanton (1983). A 7F Gaeltec catheter with two depressed microtransducers set 6 cm apart, and positioned laterally at 9 o'clock in the urethra, was used for urethral profilometry. Both resting and stress profiles were obtained. The clinical findings were not disclosed in the referral letter. The terms used and the urodynamic diagnoses made in this study conform with the definitions adopted by the International Continence Society (Bates *et al.* 1979), except for the terms 'clinical GSUI' and 'clinical DI' which are used to refer to the clinical findings defined above.

Results

Forty patients were available for study, but three patients subsequently failed to attend for urodynamic studies. The median age of the patients was 49 years (range 23–84 years). Two patients had continence surgery (one colposuspension and one vaginal repair), six had hysterectomies, and two had repair operations in the past. For convenience of presentation the patients have been divided into three groups on the basis of the presenting symptoms. Table 1 shows the clinical and urodynamic diagnoses for each of the three groups and for the entire study. The kappa statistic, which is a measure of agreement (as opposed to association) corrected for chance agreement, is given for each group.

Group 1 comprised 13 patients whose sole complaint was stress urinary incontinence. In one patient, however, this was confined to exercise, and urine loss was not demonstrable either clinically or fluoroscopically. Urine loss was demonstrable both clinically and fluoroscopically in 10 of the remaining 12 patients. The urine loss did not, however, coincide with coughing in one of these patients, but occurred after a

Table 1. Agreement between clinical and urodynamic diagnosis in 37 patients

Urodynamic diagnosis	Clinical diagnosis					Total
	Normal	GSUI	GSUI + DI	DI	UI	
Group 1						
Normal	1	2	0	0	0	3
GSUI	0	8	0	0	0	8
GSUI + DI	0	1	0	0	0	1
DI	0	0	0	0	0	0
UI	0	0	0	0	1	1
Total	1	11	0	0	1	13
Kappa = 49.4%						
Group 2						
Normal	5	0	0	0	0	5
GSUI	1	3	0	0	0	4
GSUI + DI	0	0	0	0	0	0
DI	2	1	1	0	0	4
UI	0	0	0	0	0	0
Total	8	4	1	0	0	13
Kappa = 42.5%						
Group 3						
Normal	2	0	0	0	0	2
GSUI	0	3	2	1	0	6
GSUI + DI	1	1	0	0	0	2
DI	0	0	0	1	0	1
UI	0	0	0	0	0	0
Total	3	4	2	2	0	11
Kappa = 35.3%						
All three groups						
Normal	8	2	0	0	0	10
GSUI	1	14	2	1	0	18
GSUI + DI	1	2	0	0	0	3
DI	2	1	1	1	0	5
UI	0	0	0	0	1	1
Total	12	19	3	2	1	37
Kappa = 45.8%						

GSUI, Genuine stress incontinence; DI, detrusor instability; UI, unstable urethra.

notable delay which was apparent both clinically and on fluoroscopy. A presumptive diagnosis of an unstable urethra was made, and urethral profilometry subsequently revealed uninhibited urethral relaxation not associated with incontinence. In another patient who was diagnosed as having GSUI both clinically and fluoroscopically, VCU also revealed a bladder pressure rise on standing to 22 cm H₂O. In the remaining two patients urine loss was demonstrable clinically, but fluoroscopy revealed only 'grade I stress', that is, incompetence of the bladder neck but no demonstrable urine loss. Clinical demonstration of urine loss required bladder filling in all but three of the 12 patients, and in six urine was lost only in the erect position. Interestingly, in one patient urine loss was

demonstrable only in the supine position (both clinically and fluoroscopically).

Group 2 comprised 13 patients who complained of urgency, urge incontinence, of being wet all the time or of urine simply 'coming away' at any time, but did not admit to stress incontinence. In three patients incontinence was not a prominent symptom, occurring about once per week. Three patients had GSUI, and five were normal both clinically and fluoroscopically. In one patient no abnormality was detected clinically but slight GSUI was demonstrable urodynamically. In two women urine loss was not demonstrable either clinically or fluoroscopically; bladder filling elicited severe urgency in each patient, and cystometry revealed a low compliance bladder in each case. In one patient

(A) urine loss was demonstrable both clinically and fluoroscopically. After bladder filling, the patient lost urine with the first cough, and this was followed by uncontrollable voiding. On VCU, a systolic detrusor contraction followed each cough. The coexistence of GSUI could not be satisfactorily assessed urodynamically, since urethral pressure monitoring had not been performed due to a fault in the catheter. The patient was scheduled for urethral pressure studies but failed to attend; on attempting to contact the patient it was discovered that she had died, but the cause of death was not ascertained. In the last patient (B) GSUI was demonstrable clinically in the erect position after bladder filling, but subsequent VCU showed a low pressure detrusor contraction after the third series of coughs unassociated with leakage. Neither fluoroscopy nor pressure profilometry revealed GSUI. This patient complained of incontinence premenstrually during driving, particularly over 'bumps' and while watching television. When the above tests were carried out, she was on progesterone which had produced a marked clinical improvement in her symptoms.

Group 3 comprised 11 patients who had mixed symptomatology. Six showed complete agreement between the clinical and urodynamic diagnosis: two were normal, three had GSUI, and one patient had detrusor instability. One patient was clinically normal but was found to have slight GSUI 'after much encouragement' on fluoroscopy and a low compliance bladder. Three additional patients were diagnosed as having GSUI both clinically and urodynamically: one patient was found to have urodynamically but not clinically diagnosed DI (systolic contractions) in addition to GSUI and two patients were found to have clinically (uncontrollable voiding) but not urodynamically diagnosed DI in addition to GSUI. These last two patients both

had low (<15 ml/s) urinary flow rates. One patient (C) was diagnosed urodynamically to have GSUI, but clinically she had DI. She tolerated bladder filling only to 200 ml, and remained dry in the supine position. On standing up, however, uncontrollable voiding was precipitated so that the competence of the bladder neck could not be assessed clinically.

Discussion

The present findings clearly show that, regardless of aetiology, urine loss was detected as effectively by clinical inspection as by fluoroscopy (Table 2). This finding calls into question the widespread use of fluoroscopy as a means of detecting urine loss which greatly inflates the cost of urodynamic testing. However, in only 5 of 25 patients was incontinence demonstrable clinically without bladder filling, and in only 13 of 25 was it demonstrable in the supine position. These proportions were 5 of 21 and 10 of 21, respectively, for women with GSUI, with or without coexistent DI. Therefore, a clinical examination for stress incontinence cannot be considered adequate unless the bladder is filled and the patient is examined in the upright position, and failure to ensure this must account for the previously reported insensitivity of the clinical detection of stress incontinence (Robinson & Stanton 1981; Richmond *et al.* 1986).

The ability to provoke uncontrollable voiding in some women by bladder filling was a completely unexpected finding, and probably reflected DI even though this could not always be demonstrated by cystometry. All five women in whom this sign was elicited complained of urge incontinence or of urine simply 'coming away'; DI was demonstrable cystometrically in two, and two others had low flow rates, a finding known to be associated with DI (Farrar *et al.* 1976). Furthermore, recent evidence from prolonged monitoring of bladder and rectal pressures suggests that failure to detect instability by provocative, subtraction cystometry can no longer be regarded as proof that the bladder is stable, since in some cases DI can only be identified by prolonged or telemetric monitoring (McGuire 1984; Waalwijk-Doorn *et al.* 1986; Styles *et al.* 1986). Others have observed this phenomenon either during or after urodynamic testing and interpreted the finding similarly (McGuire 1981; Mundy 1985).

The agreement between the two tests with

Table 2. Comparison of the clinical and fluoroscopic detection of urine loss in 37 patients

	Clinical incontinence		Total
	Positive	Negative	
Incontinence at fluoroscopy			
Positive	23	2	25
Negative	2	10	12
Total	25	12	37
Kappa = 75.3%			

respect to the aetiology of the incontinence was clearly not great (Table 1). Agreement was best in patients complaining only of stress incontinence (49%), but in this group disagreement between the two tests was largely due to a failure of fluoroscopy to detect two women with GSUI diagnosed clinically. It must be stressed, however, that the kappa statistic, which corrects for chance agreement, cannot be compared directly with the figures usually computed for 'agreement' between two tests (i.e. the number of women in whom the tests agreed expressed as a percentage of the total number). When calculated in this way, the values for 'agreement' between the two tests are 77%, 62%, 55% and 65% for groups 1, 2, 3, and overall, respectively (Table 1), and for urine loss the 'agreement' was 89% (Table 2).

The central, practical question is not, however, how well these two tests agreed, but the extent to which VCU improved the management of these women. In the present study the clinical and urodynamic diagnoses disagreed in 13 patients (Table 1) but this would have resulted in inappropriate treatment in only one patient (C). This patient was para 17, had a long psychiatric history, was taking antidepressants, and presented with symptoms of urgency and urge incontinence, but admitted to stress incontinence. Bladder filling elicited urgency, bladder capacity was 200 ml, urine loss was not demonstrable in the supine position, and on standing erect she promptly voided, so competence of the bladder neck could not be assessed clinically. VCU revealed GSUI and a stable bladder. She was treated with oxybutynin, and if VCU had not been performed, she would not have been offered the surgery which subsequently relieved her incontinence.

The clinical and urodynamic diagnoses were diametrically opposite in one additional patient (B) who was taking progesterone when examined. In this patient urine loss in the erect position coincided precisely with each of several coughs, yet subsequently there was no urine loss on fluoroscopy, no GSUI on UPP, and only a low pressure detrusor contraction on cystometry after the third series of coughs which was not associated with leakage. One can only surmise that DI at the time of examination had rendered the bladder neck incompetent, but continence was maintained by the distal sphincteric mechanism; coughing, however, overcame the competence of the distal sphincter. It is interesting to

speculate whether she would have voided involuntarily after bladder filling had she not been on progesterone. However, given the fact that the patient consistently denied symptoms of stress incontinence coupled with the circumstances under which urine was lost (see above), she was never considered a surgical candidate.

The present findings are clearly at variance with the observations of Jarvis *et al.* (1980) who found that 31 of 100 incontinent women would have been treated inappropriately without the information provided by a VCU. Although they do not state how their patients with combined GSUI and DI are managed, they estimated that 11 women would have undergone inappropriate surgery and 18 women with GSUI would have been treated for DI. The use of bladder filling in the present study is the most likely explanation for this difference.

Bladder filling is not, of course, being proposed as a substitute for urodynamic testing. Rather, the present findings suggest that bladder filling would be a useful adjunct to the selection of patients for further testing on clinical grounds. Surgery, without any further testing, is indicated for patients complaining solely of stress incontinence who have demonstrable stress incontinence of a significant degree. Although this is difficult to define in an objective way, the degree of incontinence is very readily apparent by inspection, as anyone who tries this method will discover. Without bladder filling, the diagnosis will not be made clinically in at least 50% of patients, and needless delay and increase in the cost of treatment incurred. Bladder filling would also provide a simple, objective and widely applicable means of assessing the result of continence surgery. Although such patients have not been investigated in the present study, there are no compelling reasons for assuming that the agreement between the clinical and fluoroscopic detection of urine loss would not apply after continence surgery.

Clearly, surgery is not indicated for women in whom urine loss is not demonstrable clinically after bladder filling. Urodynamic investigations revealed either very minor degrees of stress incontinence in one, so-called low compliance bladders in two, or both in one, or no abnormality in eight, and did not make a significant contribution to treatment. Therefore, if provocative single-channel cystometry cannot be performed in the clinic at the patient's first visit, it seems reasonable to delay referral if bladder

filling to a good volume (400–500 ml) is tolerated without any sensory urgency, and to repeat bladder filling at follow-up if the patient's initial complaint was stress incontinence, with or without frequency. Patients complaining of urgency, with or without incontinence, can also be given a trial of empirical therapy with bladder drill, oxybutinin or dicyclomine before referral, since the distinction between motor and sensory urgency or urge incontinence is not critical for initial treatment (Mundy 1985).

Whether women with mixed symptomatology and demonstrable stress incontinence require urodynamic investigations to detect those who have GSUI + DI (1 of 11 in this study) depends upon one's opinion about how such women should be treated. The traditional view has been that these women do not respond well to surgery and they may in fact be made worse. However, in two large series of patients followed for at least 3 years, McGuire *et al.* (1980) and McGuire & Savastano (1985) have found that about two-thirds of women with GSUI + DI are cured of both problems by surgery, about 10–15% have transient symptoms from DI which respond to medical treatment, and that only about 10–20% have significant residual symptoms from persistent DI. Similar short-term findings have been reported recently by other investigators (Blavais & Salinas 1985; Minini *et al.* 1985; Langer *et al.* 1986). The most disappointing surgical cure rate reported among women with GSUI + DI appears to be 47%, with most of the 'failures' reporting symptomatic improvement (Stanton *et al.* 1978; Wilkie *et al.* 1986).

Unfortunate though such failures are, one must not forget that about 17% of women with GSUI and stable bladders can be expected to develop DI *de novo* after surgery (Cardozo *et al.* 1979), and the proportion may be higher among women with GSUI who also have sensory symptoms pre-operatively (Langer *et al.* 1986). Given the relative prevalence of GSUI and GSUI + DI in this study (49% compared with 8%), one can expect that, even with a surgical cure rate of only 47% for patients with GSUI + DI, almost two-thirds of the patients who have postoperative problems referable to DI will have had stable bladders pre-operatively. Even if one takes the highest percentage reported (30%) for the frequency of GSUI + DI among women with GSUI (McGuire & Savastano 1985), one can anticipate that over 40% of patients with postoperative instability will have had stable blad-

ders pre-operatively. In other words pre-operative cystometry in the presence of GSUI is an unreliable guide to the postoperative state of the bladder in individual patients, and pre-operative cystometry by no means resolves the problem of postoperative incontinence due to DI. This is not to say that cystometry is inappropriate, but rather that it is not essential. Certainly, surgery cannot be considered inappropriate for patients in whom stress incontinence is a prominent symptom and is demonstrable clinically, and urodynamic testing is unlikely to alter their ultimate management. Exceptions would be patients with prominent symptoms of urge incontinence in which the amount of urine loss detected is slight.

By contrast, cystometry should be performed before surgery in patients who do not complain of stress incontinence or in whom this is an insignificant symptom even if on clinical examination they have GSUI. Although I consider that the one patient (B) in whom DI was confused with GSUI is very unusual and will be encountered very rarely in practice, and even more rarely, if ever, lead to inappropriate surgery, more experience is required before a decision to proceed to surgery can be based safely on the physical findings in the face of contradictory symptoms. For the same reason, urodynamic studies should also be performed in patients manifesting uncontrollable voiding particularly if this precludes an adequate clinical assessment, as in patient C, or if voiding is temporally so closely related to coughing as to render uncertain the aetiology of any antecedent urine loss which might have coincided with coughing, as in patient A.

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Neurophysiological abnormalities in genuine female stress urinary incontinence

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Summary. Perineal sensory and motor function was investigated in 28 women with genuine stress incontinence of urine and compared with a matched control group. Electrosensitivity of the dorsal nerve of the clitoris and of the urethral mucosa was significantly diminished in these patients (eight measurements 'insensitive'). Three different reflex latency measurements (dorsal nerve to external anal sphincter, dorsal nerve to urethral sphincter, urethral mucosa to external anal sphincter) were prolonged in incontinence (14 absent reflexes). Mean motor unit potential duration of the external anal sphincter was also prolonged, reflecting an early neuropathy. Anorectal manometry detected significantly weaker squeeze pressures in stress incontinence although other variables were unaffected.

Stress incontinence of urine in women is a relatively common and distressing condition. As there are often no overt neurological signs, the condition had for long been attributed to 'pelvic floor weakness', its precise aetiology remaining poorly defined. There is now some evidence for an abnormality of motor innervation of the urethral sphincter musculature in this disorder (Snooks *et al.* 1985), similar to that seen in neurogenic faecal incontinence (Henry & Swash 1985). It is conceivable that sensory abnormalities of the perineum also exist and exacerbate the urodynamic and clinical disturbances in stress urinary incontinence. These various factors may influence the results of conservative

or surgical treatment of this condition. The aim of this study was to further investigate perineal sensory and motor abnormalities in such patients using new electrophysiological techniques (Abrams *et al.* 1986).

Patients and methods

Permission for this study was obtained from the ethics committee, North Lothian District, Edinburgh on 16 June 1983, as part of a larger pelvic floor physiology project. Twenty-eight women with urodynamically proven genuine stress incontinence of urine were entered into the study. Their age range was 24-63 years (mean 41 years). Duration of symptoms ranged from 6 weeks to 23 months. Twenty-four were parous and four were nulliparous. Four dated their incontinence to a simple abdominal hysterectomy, a total of seven patients having had this operation. Three patients had previously had an anterior colporrhaphy. For purposes of comparison, 28 patients matched approximately for age and parity (age range 18-75 years, mean 44 years) with no urological symptoms were also studied. They were patients who were being

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investigated for minor colorectal and urological conditions such as polyps, bleeding, etc., and had given informed consent for this study. None of these control patients had previous pelvic floor surgery and they did not undergo urodynamic investigations. It was difficult to obtain informed consent from large numbers of incontinent and control subjects because of the 'invasive' nature of this study which included urethral catheterization, perineal electrical stimulation and the insertion of needle electrodes in the external anal sphincter. For the same reasons and due to occasional technical faults, we were not able to perform all the measurements on every subject, necessitating variation in sample sizes. However, it was ensured that the data remained comparable in terms of age and parity matching.

Urodynamics

All 28 symptomatic patients had undergone clinical examination and conventional videocystometry to ascertain the diagnosis of genuine stress urinary incontinence (Bates *et al.* 1976; Godec *et al.* 1980).

Anorectal manometry

This was performed in 11 patients using a conventional microballoon technique (Varma & Smith 1984a) to determine basal and (sustained) squeeze anal sphincter pressures, the functional sphincter length and the presence and amplitude of the rectosphincteric reflex. The rectal sensory threshold to distension was also measured (ml air) by slow inflation of the rectal balloon. It was defined as the amount of air necessary for the patient to perceive the first sensation of rectal distension and used as a measure of pelvic floor 'proprioceptive' function.

Electrophysiology

The sixth report of the International Continence Society has standardized the terminology of procedures related to neurophysiological investigation of the lower urinary tract (Abrams *et al.* 1986). The procedures used in this study are described below.

Reflex responses

The electrophysiological latency of the reflex contraction of the external anal and urethral sphincters in response to stimulation of the dorsal nerve of the clitoris was determined by a digitally averaging method. The dorsal nerve was stimulated by a felt surface electrode and the reflex responses recorded by surface ring electrodes, those within the urethra being mounted on a urinary catheter. This urethral electrode was also used as a stimulating electrode to record the urethra to anal sphincter reflex latency. The details of the methodology are as described by Smith & Varma (1984), Galloway *et al.* (1984), Fidas *et al.* (1985), and Varma *et al.* (1986). The dorsal nerve to anal sphincter reflex latency was measured (sweep 100 ms) in all the patients and controls. The dorsal nerve to urethral sphincter latency was measured (sweep 100 ms) in 15 patients and 19 controls. The urethral mucosa to anal sphincter latency was measured (sweep 200 ms) in 15 patients and 16 controls.

Electrosensitivity

Electrosensitivity of the dorsal nerve and the urethral mucosa was determined by use of the same stimulating electrodes as used for the reflex responses. The principle is based on the method described by Kieswetter (1977) and Powell & Feneley (1980). Sensory threshold was recorded in milliamperes (maximum 70 mA) and was defined as the first sensation of 'tapping' (threshold) perceived by the patient on increasing the stimulating current via the constant-current generator (DISA, Copenhagen). The dorsal nerve electrosensitivity was measured in 15 patients and 22 controls. The electrosensitivity of the urethral mucosa was determined in 16 patients and 17 control subjects.

Mean motor unit potential duration

Mean motor unit potential duration of the external anal sphincter was measured by the technique described by Varma *et al.* (1986), being a modification of the method described by Bartolo *et al.* (1983). A total of 20 motor units (10 from each side of the sphincter) were studied using a concentric needle electrode, and their mean calculated. This was used as an index of neuropathy of the external anal sphincter (Buchtal & Pinelli 1953). This investigation was per-

formed in 13 patients and 15 controls. The technique was used instead of the more sensitive single fibre electromyography because of our better acquaintance with it.

Statistics

Comparisons between control and incontinent were made by Wilcoxon's test for unpaired data.

Results

Table 1 summarizes the electrophysiological and manometric data in patients with stress incontinence and enables comparison with control patients.

Reflex response latencies

Reproducible results were obtained for all latency measurements. The three reflex response latencies were increased compared with the control group. Many of the reflexes from incontinent patients were noted to be of low amplitude (Varma *et al.* 1986). In addition, a total of 14 reflexes out of 58 were 'absent'.

Dorsal nerve and urethral mucosal electrosensitivity

Reproducible measurements were confirmed at both sites. Dorsal nerve and urethral mucosal electrosensitivity was diminished in incontinent patients (Table 1). Nine out of 31 women investigated had thresholds in excess of the maximum obtainable output (>70 mA). They were described as 'insensitive'.

External anal sphincter motor unit potential duration (MUPD)

Patients with stress urinary incontinence ($n = 13$) had prolonged MUPD of the external anal sphincter compared to control patients ($n = 15$; Table 1). This difference was significant ($P < 0.01$).

Anorectal manometry

The only significant difference that could be detected between control and stress incontinent patients by conventional anorectal manometry was a reduction in voluntary sphincter contraction pressures in the latter (Table 1).

Table 1. Electrophysiological and manometric data in stress urinary incontinence compared with controls

Variable	Stress incontinence	Control	P
DN-EAS latency (ms)	49.6 (29-70) (3 absent, $n = 28$)	39 (27.5-49) ($n = 28$)	<0.01
DN-US latency (ms)	60 (42-67) (4 absent, $n = 15$)	40.5 (30-50) ($n = 19$)	<0.01
UM-EAS latency (ms)	83 (65-120) (7 absent, $n = 15$)	69.6 (54.5-80.5) ($n = 16$)	<0.01
DN electrosensitivity (mA)	8.8 (6-14) (2 insensitive, $n = 15$)	6.1 (4-8) ($n = 22$)	<0.01
UM electrosensitivity (mA)	13.8 (9-30) (6 insensitive, $n = 15$)	8.3 (6-12) ($n = 18$)	<0.01
EAS MUPD (ms)	11.3 (8.9-15.5) ($n = 13$) ($n = 11$)	8.7 (6.2-11.1) ($n = 15$) ($n = 11$)	<0.01
HPZ (cm)	3.4 (3-4)	3.3 (3-4)	>0.1
MRP (cm H ₂ O)	93 (55-140)	108 (80-140)	>0.01
SP (cm H ₂ O)	46 (20-140)	75 (30-180)	<0.02
STV (ml air)	44 (15-140)	52 (30-100)	>0.1
Amplitude RSR (ml H ₂ O)	48 (20-90)	42 (20-60)	>0.1

All measurements show mean and range

DN, Dorsal nerve; UM, urethral mucosa, EAS, external anal sphincter; MUPD, mean motor unit potential duration; HPZ, functional anal sphincter length; MRP, maximum basal anal sphincter pressure; SP, squeeze anal sphincter pressure; STV, rectal sensory threshold volume; RSR, rectosphincteric reflex

Discussion

Genuine stress incontinence of urine is defined as the involuntary loss of urine when intravesical pressure exceeds maximum urethral pressure due to an elevation of intra-abdominal pressure and in the absence of detrusor contraction (Bates *et al.* 1976). Urethral continence at rest is dependent primarily on the bladder neck and intramural striated sphincter (external sphincter) composed of slow twitch muscle fibres which exhibit 'tonic' activity at rest (Gosling *et al.* 1981). These are supplied directly by the pelvic nerves from the sacral segments S2–S4 (Gil Vermet 1968). The periurethral striated muscle sphincter functions reflexly to augment urethral closure during 'stress' events which elevate intra-abdominal pressure such as coughing, laughing, straining, etc. This muscle receives its motor innervation from the perineal branch of the pudendal nerve (Gosling *et al.* 1981; Gosling 1985). The reflex activation of the peri-urethral musculature is mediated via excitation of its innervating α -motoneurons in the conus medullaris (S2–S4) by sensory afferents in the bladder, urethra and pelvic floor. The sensory innervation of the urethral mucosa derives predominantly from the perineal branch of the pudendal nerve (Warwick & Williams 1980). A direct branch from the motor roots of S3 and S4 reaching the pelvic floor via its visceral surface also carries sensory fibres from the posterior urethra and anal canal (Lawson 1974). The inferior rectal nerve, a direct branch of the pudendal nerve, supplies the external anal sphincter, the lining of the lower part of the anal canal and skin around the anus (Warwick & Williams 1980). Anatomical or functional deficits in the sensory, central or motor pathways of reflexes involving the urethral sphincter may, therefore, compromise urinary continence. 'Idiopathic' faecal incontinence is now recognized to be of neurogenic origin secondary to traumatic or chronic stretch injury to the innervation of the puborectalis and/or the external anal sphincter muscles. With the sophistication of electrophysiological techniques there has been accumulating evidence in favour of similar damage to the motor innervation of the striated urethral and peri-urethral muscles in stress urinary incontinence (Godec *et al.* 1980; Snooks *et al.* 1985). It is, however, conceivable that the dysfunction in such patients is further compounded by sensory deficits of the perineum.

Indeed, such an abnormality has been demonstrable in some patients with idiopathic faecal incontinence (Roe *et al.* 1986). Isolated electromyographic abnormalities of the external anal and urethral sphincters and of their motor innervation have been previously described in urinary incontinence (Fowler *et al.* 1984; Anderson 1984; Snooks *et al.* 1985). This comprehensive study has examined the perineum by a combination of electrophysiological methods to evaluate sensory impairment at two sites and reflex sphincter contraction by three different methods at both the anal and urethral sphincters. This was supplemented by direct needle EMG of the external anal sphincter and by manometric data. Reflex sphincter function is considered functionally more informative than the isolated electrophysiological abnormalities demonstrated by other methods because it mimics the 'physiological' stress response. The methods of measuring reflex response latencies used test the integrity of the entire reflex pathway and therefore have obvious advantages over other methods that measure motor latencies only or the integrity of the sphincters by needle electromyography. These other methods when used in conjunction with 'cumulative' reflex latency measurements are of value in the differential analysis of neurogenic incontinence. The additional investigation of electrosensitivity helps to examine further the nature and extent of neurological deficit. It is not possible from these results alone to incriminate 'central' neurogenic defects in Onuf's nucleus (Onuf 1901) for at least some of these abnormalities. Other studies, however, suggest that this may sometimes be the case in functional bowel and bladder disturbances (Galloway & Tainsh 1985; Vereecken *et al.* 1982; Varma & Smith 1984b; Kondo *et al.* 1986). A manometric abnormality of the external anal sphincter was demonstrable in stress urinary incontinence, although these patients had no anorectal symptoms. This is not surprising because of the larger muscle bulk of the anal sphincter, its differential innervation, and the supplementation to the maintenance of faecal continence by the substantial puborectalis muscle with its different innervation directly from the pelvic nerves (Percy *et al.* 1981). Other studies have not found a reduction in anal sphincter pressures, although they certainly exist in double incontinence (Henry & Swash 1985). These differences may simply represent the degree of neuropathy in the patient group

studied. Pelvic floor 'proprioceptive' function as determined by rectal sensory threshold appears also to be unaltered in stress incontinence. The possible adverse effects of hysterectomy on vesico-urethral dysfunction (Farghaly *et al.* 1986; Hanley 1969) may have been responsible for precipitating stress urinary incontinence in at least four patients.

This study establishes a neurogenic basis for stress urinary female incontinence. The electrophysiological methods described and their results should be of value in the management of such patients. For example, our early experience with supervised pelvic floor physiotherapy for stress incontinence suggests that those patients with severe neurological deficit do not respond to this treatment.

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Gonococcal and chlamydial antibodies in ectopic and intrauterine pregnancy

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Summary. Sera from 50 women with ectopic pregnancy and 50 age-matched control women with intrauterine pregnancy were tested by enzyme-linked immunosorbent assay (ELISA) using *C. trachomatis* and *N. gonorrhoeae* pili as antigens. In the ectopic pregnancy group 76% had IgG antibodies to *C. trachomatis* compared with 38% in the controls. The prevalence of gonococcal IgG antibody was 32% in the ectopic pregnancy group and 4% in the controls. The prevalences of IgA antibodies to the two organisms ranged from 2 to 12% and were similar in the two groups; cultures for *C. trachomatis* from cervical swabs from the ectopic pregnancy group were negative. Chlamydial antigen was detected (by ELISA) in exfoliated cervical cells of only one patient in this group.

Over recent years several countries have reported an increasing incidence of ectopic pregnancy (Beral 1975; Ory *et al.* 1981; Weström *et al.* 1981). Salpingitis is the most important predisposing factor for subsequent ectopic pregnancy (Weström *et al.* 1981), and *Chlamydia trachomatis* is the commonest sexually transmitted agent involved in salpingitis in Scandinavia (Mårdh *et al.* 1981). Numerous reports have linked salpingitis with the presence of anti-chlamydial antibodies in serum (Svensson *et al.* 1981; Mårdh & Svensson 1983; Kristensen *et al.* 1985). It is recognized, however, that many women suffer subclinical infection with *C. trachomatis* and present years later with infertility

(Moore *et al.* 1982; Conway *et al.* 1984; Brunham *et al.* 1985) or ectopic pregnancy (Brunham *et al.* 1986).

In some regions of the world gonococci are also a major cause of salpingitis, and may be expected to contribute to ectopic pregnancy. Infertility with tubal obstruction is associated with chlamydial rather than gonococcal infection (Robertson *et al.* 1987) but the relative importance of these two pathogens in ectopic pregnancy remains to be determined.

The purpose of the present study was to establish the prevalence of serum antibody to *C. trachomatis* and *N. gonorrhoeae* in women with ectopic pregnancy compared with women having normal pregnancy.

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Patients and methods

Subjects and specimen collection

During the period August 1984 to September 1986 serum was collected from 50 women with ectopic pregnancy admitted to the Princess Anne Hospital in Southampton and from 50 age-

matched women with normal pregnancy at the time of their first visit (16–18 weeks). In addition, two cervical swabs were taken from the ectopic pregnancy group for culture of *C. trachomatis* and for detection of *C. trachomatis* antigen by ELISA (antigen trapping). Samples of fallopian tube were collected at surgery. Sera and tissue were stored at -20°C and swabs at -70°C .

Clinical details were obtained from the ectopic pregnancy group with regard to previous history of salpingitis, subfertility and contraceptive methods.

Antibody detection

Conventional ELISA for serum IgG and IgA was performed on quadruplicate 1/100 dilutions of sera, using *C. trachomatis* L1 and *N. gonorrhoeae* P9–2 pili as antigens (Mabey *et al.* 1985). Peroxidase conjugates were from Dako, Copenhagen, Denmark. Chlamydial culture transport fluid was tested similarly for transudated IgG and IgA. Quadruplicate 1/100 serum dilutions were used, rather than end-point titration, for greater convenience of assay, economy of antigen and reduced standard error. Excellent correlation between maximum optical density (OD 450 nm) and titre end-point has been shown previously (Robertson *et al.* 1987).

Culture of *C. trachomatis* and detection of chlamydial antigen

Culture swab material was centrifuged onto cycloheximide-treated BGMK cells on coverslips and incubated for 48 h (Ripa & Mårdh 1977). Coverslips were reacted with rabbit anti-*C. trachomatis* LGV L1 antiserum, followed by fluorescent swine anti-rabbit serum (Dako, Copenhagen, Denmark).

C. trachomatis antigen was detected on cervical swabs using the Boots Celltech IDEIA Kit, following the manufacturer's instructions. In addition, specimens of fallopian tube were boiled in IDEIA Kit transport medium, centrifuged, and the supernatants tested for *C. trachomatis* antigen.

Culture of *N. gonorrhoeae* from cervical swabs was not performed.

Results

During the 2-year period of specimen collection a total of 74 women were admitted with ectopic pregnancy. Our sample of 50/74 women gave 67% representation of this group.

Prevalence of antibodies to *C. trachomatis* and gonococcal pili

Figure 1 shows the frequency distribution of IgG and IgA antibodies to *C. trachomatis* and *N. gonorrhoeae* pili in the women with ectopic pregnancy and in the control group with intrauterine pregnancy. On the basis of these distributions an optical density (OD) 450 nm ≥ 0.3 was considered positive for IgG antibodies to *C. trachomatis*, and OD 450 nm ≥ 0.5 for gonococcal pili. For IgA antibodies an OD 450 nm ≥ 0.4 was considered positive for both antigens. Of the women with ectopic pregnancy 76% (38 of 50) were positive for IgG antibody to *C. trachomatis* L1 compared with 38% (19 of 50) in the control group (Table 1). For IgG antibodies to gonococcal pili, 32% (16 of 50) of the women with ectopic pregnancy were positive compared with 4% (2 of 50) in the controls. When infections with the two organisms were considered together, total infection in the ectopic group was 86% with 22% having antibodies to both organisms. In the control group 40% had been infected, only one woman having antibodies to both organisms.

The distribution of serum IgA antibodies to the two organisms is shown in Fig. 1. In the ectopic pregnancy group 12% (6 of 50) patients were positive for *C. trachomatis* antibody compared with 2% (1 of 50) in the controls. All these patients also had high levels of IgG antibody, suggesting that these were past infections. No difference was found in the two groups in prevalence of serum IgA antibody to gonococcal pili: 4% (2 of 50) were positive in each case. Two of the four ectopic pregnancy sera positive for IgA antibodies against gonococcal pili did not have correspondingly high IgG levels, suggesting that their infection may have been recent.

Antigen detection

Chlamydial antigen was detected by ELISA in cervical swabs from only one patient in the ectopic pregnancy group. This patient had a high level of serum IgG antibodies to *C. trachomatis* and borderline IgA antibodies. No chlamydiae

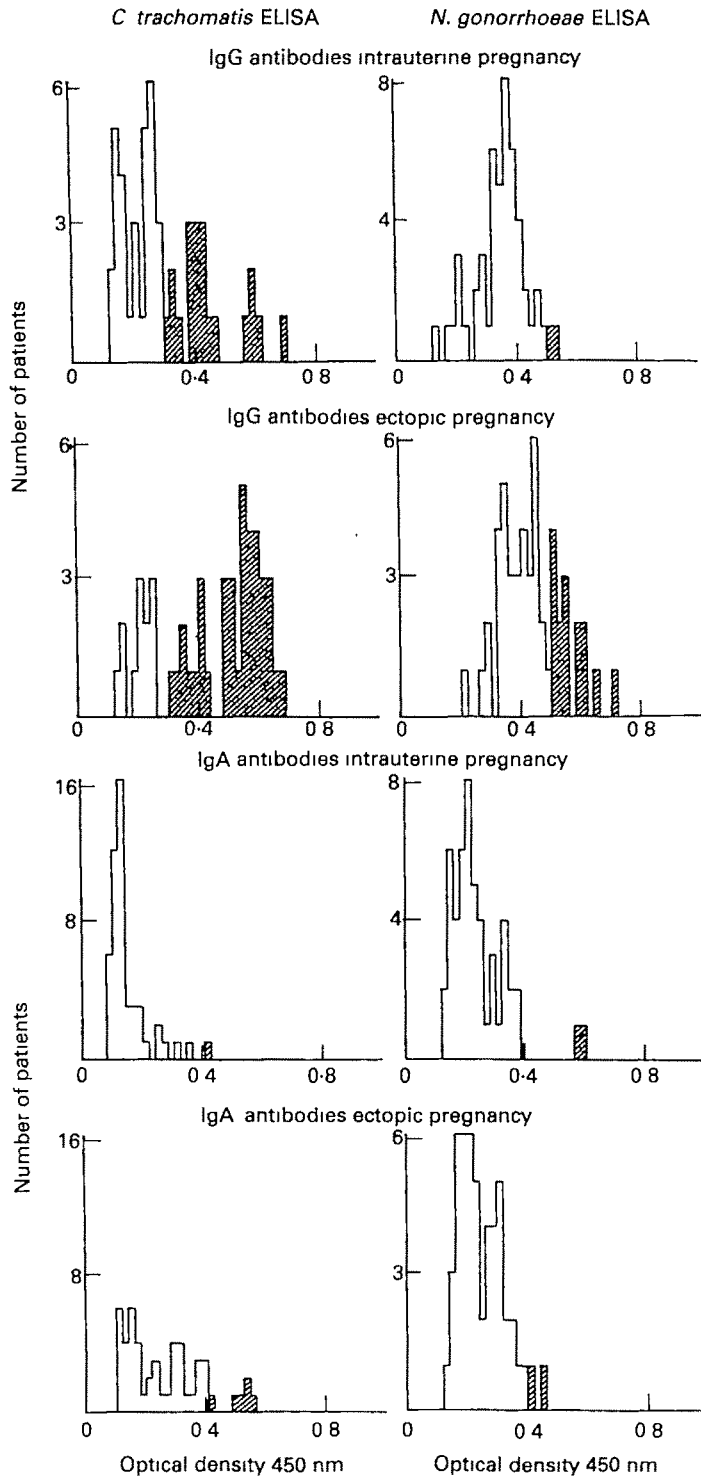


Fig. 1. Frequency distribution of IgG and IgA antibodies to *C. trachomatis* L1 and *N. gonorrhoeae* P9-2 pili in women with ectopic pregnancy and intrauterine pregnancy. ELISA OD 450 nm considered positive have been shaded.

Table 1. Prevalence of IgG and IgA antibodies to *Chlamydia trachomatis* L1 and *Neisseria gonorrhoeae* P9 pili by ELISA in women with ectopic and intrauterine pregnancy

Antibody	Number (%) of positive sera		χ^2 *	P*
	Intrauterine pregnancy (n = 53)	Ectopic pregnancy (n = 50)		
IgG				
<i>C. trachomatis</i>	19 (36)	38 (76)	13.21	<0.001
<i>N. gonorrhoeae</i> pili	2 (4)	16 (32)	11.44	0.001
IgA				
<i>C. trachomatis</i>	1 (2)	6 (12)	Not significant	
<i>N. gonorrhoeae</i> pili	2 (4)	2 (4)		

*Significance in χ^2 -test with Yates' continuity correction factor of difference between patients with ectopic pregnancy and intrauterine controls.

were isolated from any of these patients and no chlamydial antigen was detected in any fallopian tube tissue.

ELISA of cervical swab fluid

Cervical swab fluid from the ectopic pregnancy group was tested for IgG and IgA antibodies to *C. trachomatis* and gonococcal pili. Cervical IgG antibodies to *C. trachomatis* were found in 55% (18 of 33) and IgA antibodies in 43% (13 of 30). Only one patient (3%) had cervical IgG or IgA antibodies to gonococcal pili.

Clinical histories

Of the 50 women with ectopic pregnancy only three gave a history of salpingitis, but 18 gave a history of subfertility although not all had consulted a doctor. Of these 21 women 17 were positive for *C. trachomatis* antibody (81%) and eight (38%) for antibody to gonococcal pili, compared with 72% and 28% respectively for women with no clinical history. The mean ELISA OD 450 nm for *C. trachomatis* antibody was 0.48 for those with a clinical history and 0.39 for those without.

Twelve women with ectopic pregnancy had used an intrauterine contraceptive device (IUCD) previously and had a lower prevalence of serum antibody to *C. trachomatis* (58%) or to *N. gonorrhoeae* pili (17%) than those who were non-users (82% and 37% respectively). The mean ELISA OD 450 nm for *C. trachomatis* in the IUCD users was 0.39 compared with 0.46 for the non-users. Tests of statistical significance

were not performed on these data owing to the small sample size.

Discussion

The group of 50 women with ectopic pregnancy had significantly more serum IgG antibodies to *C. trachomatis* and *N. gonorrhoeae* pili than the 50 control women with intrauterine pregnancies. An association between chlamydial antibodies and ectopic pregnancy has also been reported from Sweden (Svensson *et al.* 1985), Finland (Puolakkainen *et al.* 1986) and Canada (Brunham *et al.* 1986) but not from the UK. An association between ectopic pregnancy and gonococcal antibodies has not been reported previously.

The 38% prevalence of *C. trachomatis* serum IgG antibody in the control group confirms previous findings in the UK (Conway *et al.* 1984; Robertson *et al.* 1987). It is not clear to what extent this reflects previous exposure to sexually transmitted disease as opposed to cross-reactivity with antibody to other agents such as the newly described TWAR strains of *C. psittaci* (Forsey *et al.* 1986). The finding of significant amounts of antibody to gonococcal pili in the ectopic pregnancy group is striking when compared to our previous study on subfertile women in Bristol where serological evidence of past infection with gonococci was not found to be an important factor in tubal damage (Robertson *et al.* 1987). In that study only women seeking termination of pregnancy had significant evidence of gonococcal infection. In other regions of the world gonococci play a major role in subfertility due to tubal damage (Mabey *et al.* 1985) but we

know of no obvious socio-economic factors in the study populations which might explain the increased prevalence of gonococcal pilus antibodies in Southampton women suffering ectopic pregnancy compared with Bristol women with infertility due to tubal obstruction. One may speculate that subtle differences in the pathogenicity of these two organisms for the fallopian tube could explain these observations. Each organism gives rise to obstruction of the fallopian tube (infertility) as a result of fibrosis and scarring following acute inflammation. Scarring distortion of the tarsal plate is a feature of chronic chlamydial ocular infection (trachoma) whilst urethral stricture was a common complication of chronic gonococcal infection in the pre-antibiotic era. In addition, gonococci cause marked exfoliation of ciliated epithelia from fallopian tube organ cultures (Ward *et al.* 1974) suggesting that they may alter egg transport in the non-obstructed fallopian tube giving rise to ectopic pregnancy.

There was no evidence of recent infection in the ectopic pregnancy group or control group; high serum IgA levels were always associated with high levels of IgG. Similarly, high IgA levels in cervical swab fluid from ectopic pregnancy patients correlated with high IgG. Our failure to detect *C. trachomatis* antigen or isolates in all but one of the cervical swabs or in tissue in the ectopic group also supports the hypothesis that the infections had resolved by the time the ectopic pregnancy became apparent. There would be thus no benefit in instituting treatment in these patients.

Twelve (24%) of the ectopic pregnancy group gave a history of IUCD usage. This group had lower antibody levels to *C. trachomatis* than those not using an IUCD, a phenomenon also noted by Svensson *et al.* (1985) and Brunham *et al.* (1986). Paradoxically, the acute inflammatory response stimulated by the insertion of an IUCD might help eliminate chlamydial infection in the upper genital tract.

The results reported here suggest that both gonococci and chlamydiae are major causes of ectopic pregnancy in the Southampton area. Most of these infections are subclinical, since only three women gave a history of salpingitis, and have resolved before the condition manifests. Further studies are needed to establish whether the differences we have observed in the prevalence of gonococcal infection in subfertility

and ectopic pregnancy are true in other areas of the country and, if so, its pathological basis.

Acknowledgment

We are grateful to Dr John Heckels and his group for the kind gift of purified gonococcal pili.

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Pregnancy in women with thalidomide-induced disabilities. Case report and a questionnaire study

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Case report

Pregnancy in a patient with thalidomide-induced four-limb disability

This 24-year-old woman with congenital upper limb amelia and lower limb phocomelia, as a result of intrauterine exposure to thalidomide, was pregnant for the first time. She was living in a residential home for the disabled, being wheelchair bound and needing much physical help with everyday activities. Her 37-year-old husband was disabled by multiple sclerosis; he had four children from a previous marriage. At the first antenatal clinic visit she proved to be an intelligent woman with a strong personality, who was obviously determined to be as independent as possible. She had had a laparotomy for suspected appendicitis, a tonsillectomy and adenoidectomy, and an operation to widen her left ear canal. She had complete absence of the upper limbs and rudimentary lower limbs. Systematic examination was otherwise normal. For practical reasons it was impossible at the first visit to weigh her or estimate her blood pressure, but soon afterwards she was found to weigh 64 kg and to be normotensive. An ultrasound scan revealed an intrauterine pregnancy compatible with 12 weeks of amenorrhoea.

Haematological investigations were deferred as no peripheral veins suitable for venepuncture could be found. Subsequent investigations were performed using small quantities of blood

obtained from ear-lobe punctures, and showed her to be rhesus negative.

Her management was planned at a case conference which was attended by all the staff likely to be involved in her care.

She was transferred from her wheelchair to the examination couch in the antenatal clinic with a canvas sling attached to a hoist. Even with this appliance, two people were needed to help with lifting. Weighing was organized at her home for the disabled, where suitable facilities existed. To avoid difficulties with transport some of her antenatal care was provided by a community midwife. Her temporal arterial blood pressure was estimated with the aid of a paediatric cuff and a ruler. The cuff was inflated and one end was tied off whilst the other was attached to an ordinary mercury manometer. This cuff was then placed over the temporal artery in front of the ear and a ruler placed over it. Graduated finger pressure on the ruler applied by an assistant was used to occlude and gradually decompress the temporal artery. Simultaneous auscultation over the temporal artery distal to the occlusion detected the systolic and diastolic blood pressures. The difference in height (in cm) between the temporal artery and the angle of Lewis was added to the blood pressure reading to obtain the corrected blood pressure.

At 25 weeks gestation she was treated in hospital for 2 days with ampicillin for a urinary tract infection. Her haemoglobin level was 9.4 g/dl in spite of prophylactic iron but it gradually increased to 11.9 g/dl by 36 weeks gestation. From 35 weeks gestation she was an in-patient because of her anxiety about backache and uterine tightenings.

Fetal growth, monitored both clinically and by regular ultrasound scans, was satisfactory. We were able to reassure the patient about the normality of her fetus, and in particular were able to demonstrate the fetal limbs.

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At 37 weeks gestation CT scan pelvimetry showed the anteroposterior diameter of the pelvic inlet and outlet to be 12.6 cm and 10 cm respectively. Transverse measurements were not obtained due to the patient's obesity and osteopenia. During the 40th week the membranes ruptured spontaneously. An intravenous jugular catheter was inserted and labour was augmented with an intravenous oxytocin infusion. A healthy female infant weighing 3.4 kg was born by caesarean section because of failure to progress in the first stage after 9 h of regular, strong uterine contractions. During the operation which was technically difficult due to the patient's obesity, a left rudimentary uterine horn was noted which did not appear to communicate with the uterine cavity proper. There were no other abnormalities.

Postnatally the haemoglobin level dropped to 6.6 g/dl and the patient was transfused with four units of blood. She also developed a post-operative wound infection.

The baby was bottle fed. Both mother and baby were discharged from hospital on the 19th postnatal day to their specially adapted home. The local Social Services Department, in association with the Thalidomide Trust, were instrumental in organizing suitable long-term accommodation and support for the couple, their newborn baby and for two of the children from the husband's previous marriage. This support involved one full-time trained nurse helping at home.

At the postnatal check mother and baby were both well. A combined oral contraceptive pill was prescribed.

Questionnaire study

We circulated a questionnaire via the United Kingdom Thalidomide Trust to all their benefici-

aries who had given birth. The women were asked to send details of their disabilities and their pregnancies and delivery, including any complications they could remember. No specific complications were suggested. Forty-nine questionnaires were sent and we received 34 replies and added our one further personal case producing a response rate of 67% to the questionnaire. These 35 women had 64 livebirths and six miscarriages between them, a miscarriage rate of 9%. There were no reports of abnormality amongst the 64 livebirths. Most of these women had minor disabilities of the upper limbs and ears, but there were six women with absent upper limbs and two women with absent lower limbs, including our patient in whom all four limbs were missing. There was one woman with minor upper limb disability who also had a congenitally dislocated hip. Details provided by the Thalidomide Trust showed that all the women who had whole limbs missing had responded to the questionnaire.

The complications of pregnancy included pre-eclampsia in four, premature labour in three, urinary tract infection in two, twin pregnancy in one and retained placenta in one.

Of the 64 deliveries, 46 (72%) were normal, 4 (6%) were forceps deliveries and 14 (22%) were by caesarean section. The average birthweight was 3.6 kg (range 1.4–4.1 kg).

Further analysis of the mode of delivery in relation to the degree of disability (Table 1) shows that the two women with absent lower limbs had all their deliveries (four in total) by caesarean section. Three of the remaining five women with absent upper limbs had five abdominal and no vaginal deliveries between them. Only two women out of the six with the absent upper limbs had vaginal deliveries. In the group of 28 women with minor disabilities, there were five caesarean sections, one of which was due to an abnormally formed pelvis due to congenital hip dislocation; out of a total of 52 deliveries, the caesarean section rate was 10%.

Difficulties were experienced by the obstetric staff in measuring the pulse rate and blood pressure, and difficulties were experienced by the mothers in feeding and changing the babies. The use of a sling to support the baby on the mother's chest was helpful. Episiotomy pain restricted the use of the mother's feet which quite often are a substitute for missing or malformed hands or arms.

Table 1. Degree of disability and caesarean section rate

	No. of women	No. of deliveries	Caesarean sections
			<i>n</i> (%)
Absent lower limbs	2*	4	4 (100)
Absent upper limbs	6*	9	6 (67)
Minor disabilities†	27	51	4 (8)†

* One woman with all limbs absent, included twice.

† Excluding the woman with congenital hip dislocation

Discussion

The first suggestion that thalidomide might be teratogenic in man came from West Germany (Lenz 1962; Pfeiffer & Kosenow 1962), and Australia (McBride 1961). The malformations attributed to this drug include phocomelia, radial hemimelia, microtia, intestinal atresia, heart lesions, renal anomalies, hypoplasia of the gall bladder and appendix, and facial naevus (Newman 1986). The total number of thalidomide-affected children in the UK is about 450, all with disabilities ranging from severe incapacity to no disability at all (Smithells 1973; Quibell 1981). In the group with four-limb phocomelia, the disability is obviously extreme.

Gynaecological abnormalities have been reported previously in only five of the British patients (Quibell 1981), but full examinations were not undertaken at the time of the assessments as these were considered unethical. Now that the affected children are older, more reproductive tract abnormalities are being discovered (Hoffman *et al.* 1976). Spinal abnormalities have been underestimated as the anterior fusion of the vertebrae can be diagnosed only by a lateral X-ray of the spine and this was done only in a proportion of the children (Quibell 1981). These fusions are often extensive in the dorso-lumbar area. Other anomalies include scoliosis, kyphosis, sacral agenesis and spondylolisthesis. When the bony pelvis is deformed, or when there are anterior fusions of the vertebrae in the lumbosacral region, parturition may be difficult and caesarean section may be necessary.

With our patient, we were particularly concerned about pelvic shape and size, not only of congenital origin, but also secondary to the lack of the normal weight-bearing stresses that promote bone growth. The radiologists felt that CT scan pelvimetry would be technically easier and more helpful than the conventional X-ray pelvimetry. It also had the advantage of reducing the radiation dose from 1 mGy to 0.1–0.2 mGy (Suramo *et al.* 1984). The decision to allow a trial of labour was based on these measurements and the clinical findings.

Only four of the women in the survey were found to be anaemic in pregnancy and our patient was the only one with all four limbs absent. Women with absence of some limbs, such as bilateral amputees, maintain their haemoglobin levels within the normal range (I. Burton, personal communication).

This unusual pregnancy, in a woman with extreme disability, presented us with several taxing practical problems. In overcoming these problems we were greatly assisted by the woman's determination to be as independent as possible. We were very impressed by her capacity to cope with her disability and the extra burden imposed by the pregnancy.

In the questionnaire study, we found a high caesarean section rate in the women with major malformations, especially of the lower limbs. This reinforces the view that mechanical difficulty in labour is the main obstetric problem in this group; otherwise, the overall reproductive performance of the women with thalidomide-induced disabilities is good. It is worth noting that all the children born to mothers who replied to the questionnaire were physically normal, supporting the reassurance given by Newman (1986) that there is no increased risk of congenital abnormality among the offspring of thalidomide victims.

Acknowledgments

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Granular cell myoblastoma of the cervix. Case report

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Case report

A 55-year-old woman (gravida 4, para 4) was admitted to the gynaecological ward of Golda Meir Medical Center following an episode of postmenopausal bleeding and lower abdominal pain.

Previous history revealed hypertension of 10 years duration, which had been treated by beta-blocking agents.

The routine twice-yearly gynaecological examinations during the menopausal period (since the age of 50) were unremarkable.

On admission her general condition was satisfactory. The gynaecological examination demonstrated a polyp, measuring 0.5×0.5 cm, on the anterior lip of the uterine cervix. The uterus was within normal shape and size. Both adnexa were normal.

Laboratory findings, including blood count and chemistry, liver and kidney function were within normal limits. Chest radiograph was normal.

Under general anaesthesia, cervical polypectomy and fractionated dilatation and curettage were performed. No material was found in the cervical canal or uterine cavity.

The postoperative course was uneventful and the patient was discharged a few hours after the procedure.

A follow-up of 3 years did not reveal any other abnormality.

Histological examination revealed a polypoid lesion covered by flat exocervical mucosa (Fig. 1). In the stroma there were some nabothian cysts and a small granular cell myoblastoma. It was composed of closely packed large cells with granular cytoplasm and small centrally located nucleus (Fig. 2). The granules were PAS positive. The peroxidase antiperoxidase method of Sternberger (1979) was used with commercially available kits (DAKO) to identify S-100 related antigen and this was positive in the cytoplasm of the granular cell myoblastoma.

Discussion

Granular cell myoblastoma of the tongue was first described by Weber (1854) and then by Abrikossoff (1926) who believed that the tumour was derived from striated muscle cells. Since then identical lesions have been described in almost every tissue of the body, including the stomach (Gal *et al.* 1985), bladder (Fletcher *et al.* 1985), penis (Stone *et al.* 1983) and breast (Ingram *et al.* 1984). However, the female reproductive system has rarely been involved; we have found no reports of granular cell myoblastoma in the fallopian tubes, uterus or ovaries and only one report of this tumour in the uterine cervix by Copas *et al.* (1981). In contrast the vulvar area is a site of predilection (Dgani *et al.* 1978).

On clinical examination the granular cell myoblastoma presents as a solitary, painless mass, resembling a sebaceous cyst. Multiple tumours are encountered in 10% of the cases (McSwain *et al.* 1980), so that extragenital sites should always be examined.

Excisional biopsy is the treatment of choice. The patient should be followed up since recurrence is possible. Malignant varieties of granular cell myoblastoma are rare (von Mägori & Szegvári 1973).

One of the remarkable features of the tumour is the epidermal acanthosis that overlies the lesion and may simulate squamous cell car-

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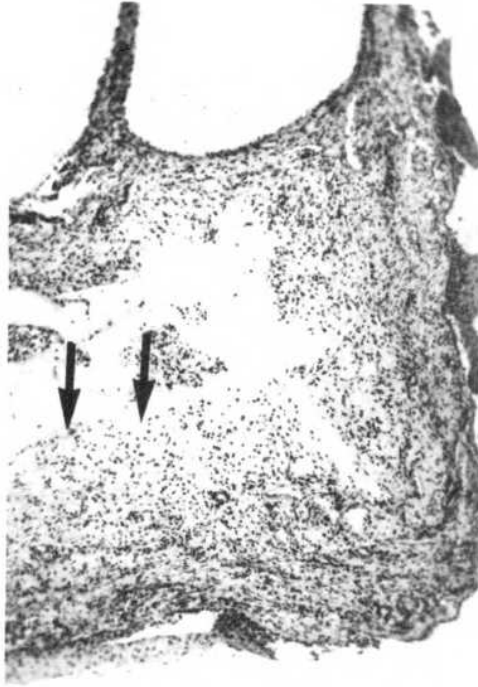


Fig. 1. Cervical polyp with granular cell myoblastoma (arrow). Haematoxylin and eosin. $\times 20$.

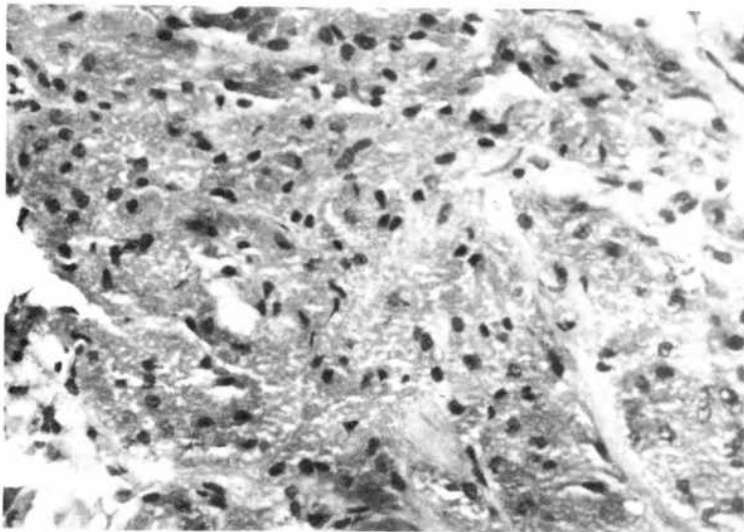


Fig. 2. Section of the granular cell myoblastoma showing large cells with centrally located small nucleus and granular cytoplasm. Haematoxylin and eosin. $\times 340$.

cinoma. In our patient the myoblastoma did not provoke any changes in the endocervical mucosa. Nevertheless, the pathologist must always be aware that epithelial proliferation may be caused by an underlying granular cell myoblastoma.

Feyrter (1935) was the first to suggest that this tumour was of neural origin. The presence of neurone-specific enolase (Rode *et al.* 1982), and the presence of S-100 protein in granular cell myoblastoma (Stefanson & Wollmann 1982) and also in our patient strongly suggests a Schwann cell origin.

The cervical granular cell myoblastoma in our patient did not differ histologically or immunohistochemically from this tumour at other locations.

The predilection of this tumour for the tongue and the skin in contrast to its rare occurrence in the female genital tract is still unexplained.

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Uteroplacental and umbilical artery blood velocity waveforms in placental abruption assessed by Doppler ultrasound. Case report

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Case report

A 39-year-old para 1 with mild pre-eclampsia (blood pressure 140/90 mmHg) was referred for ultrasound assessment because of painless uterine contractions and a decelerative fetal heart rate pattern on cardiotocogram at 31 weeks gestation. There was uterine relaxation between contractions and no vaginal bleeding. Ultrasonic scanning revealed a single fetus of estimated weight 1100 g with normal anatomy. The placenta was situated at the fundus and a large retroplacental clot was noted.

The uterine and umbilical artery velocity waveforms were assessed using a continuous-wave Doppler system (Multigon, New York) between uterine contractions under direct ultrasound visualization. The umbilical artery exhibited a systolic/diastolic (A/B) ratio of 8 (normal <3.7) (Trudinger *et al.* 1985) while that in the uteroplacental vessels was 6 (normal <2.6) (Fleischer *et al.* 1986) (Figs 1 & 2). The fetal heart rate was 120 beats/min at the time of recording. In view of the placental abruption, immediate caesarean section was performed and a live female infant weighing 1130 g was born. A fresh retroplacental clot (volume approximately 500 ml) was identified at delivery.

Comment

Large placental abruption is often associated with sudden fetal death although the mechanism

is uncertain (Sholl 1987). In our patient a major degree of placental separation was fortuitously observed during an ultrasound examination and Doppler flow studies were performed. The velocity changes in response to abruption are dramatic and have not been reported previously.

The flow velocity waveform obtained using Doppler ultrasound is believed to reflect changes in vascular resistance (Trudinger *et al.* 1985). The findings in this patient suggest a large resistance to blood flow in both the maternal and fetal placental circulations in response to placental separation. A diastolic notch was seen in the umbilical artery waveform, which is an exceptionally rare finding in an umbilical artery sonogram. A notch is more commonly found in the uterine artery where it is associated with increased vascular resistance (Griffin *et al.* 1983).

The cause of a notch in the umbilical artery sonogram is uncertain but the retroplacental clot may cause a tamponade effect thus increasing resistance in the fetoplacental vascular bed. Alternatively, release of prostaglandins by placental separation has a powerful vasoconstrictor effect and may cause vasospasm in the small vessels of both the uterine and umbilical circulations. This may account for the changes seen in both maternal and fetal vessels in this case.

It is postulated that this sudden increase in vascular resistance may be the cause of fetal death in some patients with placental abruption where there is apparently insufficient placental separation to account for death due to hypoxia or fetal haemorrhage.

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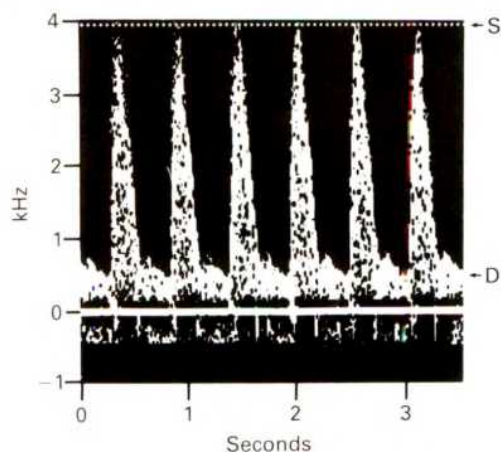


Fig. 1. The umbilical artery sonogram. The A/B ratio is 8 (normal <3.7) and there is a diastolic notch present in early diastole.

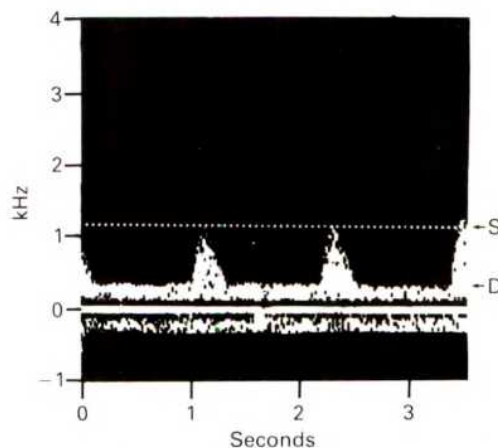


Fig. 2. The uterine artery sonogram. The A/B ratio is 6 (normal <2.6).

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BLAIR BELL RESEARCH SOCIETY

Papers presented at the meeting on 21 January 1988 at the Royal College of Obstetricians and Gynaecologists

The effect of anti-sperm antibodies on the fertilizing capacity of men undergoing immunosuppressive therapy by J. E. Swann & R. W. Shaw. Academic Department of Obstetrics and Gynaecology, Royal Free Hospital, London NW3 2QG

The presence of anti-sperm antibodies (ASAB) has been associated with reduced fertility in men. The detrimental effects of ASAB on sperm transport have been well demonstrated, and evidence in recent years also indicates interference in the process of sperm-egg interaction. Immunosuppressive treatment has produced varying degrees of success in this condition. An attempt is made to assess the effect of ASAB on the fertilizing capacity and the efficacy of immunosuppressive therapy. The semen of 31 men exhibiting elevated serum anti-sperm antibody levels as measured by the tray agglutination technique (TAT) were tested with the hamster egg penetration test (HEPT). Antibody titres of 1:32 or greater were shown to inhibit sperm penetration in 21 cases (67.7%). In the remaining 10 cases (32.3%) positive penetration was recorded. This contrasted significantly with the results from 137 infertile men where ASAB had been excluded giving 78.1% positive, 21.9% negative ($\chi^2 = 23.0$, $P < 0.001$). Twenty men with negative HEPT were again examined after 3 months of prednisolone therapy (5 mg three times daily) and assessed for egg penetration and anti-sperm antibody levels. Steroid treatment was found to improve HEPT performance in 14 cases (70.0%). In the steroid-treated group 13 men also took part in in-vitro fertilization (IVF) on the basis of their partner's clinical assessment. In seven of the men, the semen showed improved HEPT and fertilizing capacity at IVF. This study indicates that ASAB inhibit sperm-egg interaction in most cases although positive results in spite of ASAB can possibly be attributed to their polyclonal nature. Also

demonstrated is an improved prognosis for fertility using immunosuppressive therapy where sperm transport is circumvented by IVF.

A preliminary study of the serum lipid profiles of patients undergoing therapy for endometriosis by R. L. Gardner, D. Crook* & R. W. Shaw. Academic Department of Obstetrics and Gynaecology, Royal Free Hospital, London and *The Cavendish Clinic, London NW8

Twenty-five women aged 18-40 years (mean 32.4) have been randomly allocated Zoladex depot (ICI 118,630, goserelin, ICI Pharmaceuticals, Macclesfield) by subcutaneous injection, 3.6 mg monthly, or oral danazol (Winthrop Laboratories, Surrey) 600 mg daily, as treatment for their endometriosis. A pretreatment sample of blood was collected after a 6-h fast for lipid estimations on EDTA plasma. These assays were repeated every 2 months during the treatment period. Oestradiol levels were measured before treatment and monthly during therapy. After 4 months of therapy, eight patients treated with danazol had a significant rise in LDL cholesterol from 91.1 (SE 9.4) to 128.8 (SE 14.4) mg/dl ($P < 0.01$) and its carrier protein apolipoprotein B from 57.0 (SE 4.6) to 82.9 (SE 9.4) mg/dl ($P < 0.01$). A significant fall was observed in total HDL from 60.9 (SE 3.7) to 28.8 (SE 2.6) mg/dl ($P < 0.01$) and HDL2 from 14.1 (SE 2.3) to 1.5 (SE 0.5) mg/dl ($P < 0.01$). These changes, which are similar to those found in individuals who are at risk of developing ischaemic heart disease, may be important in those danazol-treated women who may be in a high-risk group of developing ischaemic heart disease or where danazol therapy is prolonged. In the 17 women treated with Zoladex there were no significant changes in the lipid profile which suggests that Zoladex depot does not promote atheroma formation and the development of ischaemic heart disease. Both danazol and

Zoladex depot produced a state of hypo-oestrogenism so the different lipid profiles during treatment cannot be due entirely to hypo-oestrogenism alone.

The assessment of the role of CA 125 in the diagnosis, prognosis and efficacy of treatment in endometriosis by L. M. Irvine, G. Burford, P. Acien*, & R. W. Shaw. Academic Department of Obstetrics and Gynaecology, Royal Free Hospital, Pond Street, London NW3 2QG and *Department of Obstetrics and Gynaecology, University of Alicante, Spain

The level of CA 125, an antigen defined by the monoclonal antibody OC-125, is associated with a high molecular weight serum glycoprotein and is raised in a number of conditions including endometriosis. We measured serum CA 125 in women suffering from endometriosis and asked the following questions: Is there a difference in serum CA 125 levels between normal women and those with a laparoscopic diagnosis of endometriosis? Is there a correlation between serum CA 125 level and the severity of the endometriosis as scored using the American Fertility Society system? Does the level of CA 125 fall with treatment of endometriosis? We compared serum CA 125 levels in 30 normal non-menstruating women with 58 women with endometriosis diagnosed laparoscopically undergoing treatment with danazol (Winthrop Laboratories, Surrey) 200 mg three times daily ($n=18$), Buserelin (Hoechst UK Ltd, Middlesex) 300 µg three or four times daily by nasal spray ($n=11$), Nafarelin (Syntax Pharmaceuticals, Berkshire) 200 µg twice daily by nasal spray ($n=10$) or Zoladex (ICI Pharmaceuticals, Cheshire) 3.6 mg by subcutaneous injection monthly ($n=19$). Serum CA 125 was measured using an immunoradiometric assay kit (Abbott Diagnostic, Wokingham, Berkshire). Mean serum CA 125 level in normal women was 11.8 (SE 1.6) while the level in the endometriosis group was 42.9 (SE 8.5) u/ml ($P<0.01$). After 6 months of treatment the mean serum CA 125 level had fallen to 15.0 (SE 1.7) u/ml in the endometriosis group, not significantly different from that in the controls. We found no association between CA 125 levels before treatment and severity of the endometriosis, as assessed by the American Fertility Society scoring system. Two months after the therapy had stopped CA

125 levels, although rising, had not reached pre-treatment levels and none of the patients had evidence of a recurrence at that time. Whether CA 125 levels can be used to predict the presence of endometriosis or its recurrence needs to be investigated.

Pregnancy-associated endometrial α_2 -globulin (α_2 -PEG): assessment as a potential marker for endometrial adenocarcinoma by P. L. Wood*, G. T. Waites*, A. C. Davidson*, R. A. Walker† & S. C. Bell*‡. Departments of *Obstetrics and Gynaecology, †Pathology and ‡Biochemistry, University of Leicester, Leicester LE2 7LX

Pregnancy-associated endometrial α_2 -globulin (α_2 -PEG) has been demonstrated to represent the quantitatively major secretory product of the normal endometrium in the mid to late luteal phase of the cycle, being localized to the glandular epithelium. It is as yet unknown whether endometrial adenocarcinoma produces the protein or is stimulated to produce it by the administration of oral progestogens. The recognition of such production would have the potential for the protein to serve as a marker for endometrial adenocarcinoma and progestogen responsiveness. Endometrium from 16 postmenopausal women with endometrial adenocarcinoma was examined immunohistochemically using a monoclonal antibody to α_2 -PEG. Tissue was examined at the time of diagnosis and the time of definitive therapy following oral progestogens. Regardless of tumour differentiation, malignant endometrium does not appear to produce α_2 -PEG. It was noted that adjacent non-malignant endometrium responded dramatically to oral progestogen with the appearance within the glandular epithelium of immunoreactive α_2 -PEG. α_2 -PEG does not appear to be a suitable marker for endometrial adenocarcinoma but its production in non-malignant postmenopausal endometrium can be stimulated by oral progestogens, suggesting that α_2 -PEG may be a useful marker of relative progestogenic potency.

Reduction of uterine arterial blood flow, as assessed by Doppler ultrasound, in patients with leiomyoma treated with GnRH agonists by W. H. Matta, I. Stabile*, R. W. Shaw & S. Campbell*. Academic Departments of Obstetrics and

Gynaecology at Royal Free Hospital, London NW3 2QG and *King's College Hospital, London SE5 9RS

Chronic administration of gonadotrophin-releasing hormone (GnRH) agonists can significantly reduce the size of uterine leiomyomas. It is possible that this reduction in volume could be related, in part, to induced changes in uterine blood flow. We studied impedance to blood flow within the uterine vasculature by Doppler ultrasound (pulsed wave duplex system) in eight patients, aged 26–41 years, with abdominally palpable uterine leiomyomas. Each patient received 400 µg of intranasal buserelin [D-Ser (TBU)⁶des-Gly-NH₂¹⁰ LHRH ethylamide] (Hoechst Pharmaceuticals), three times daily for 4 months. Pretreatment and monthly assessments were made of serum 17-β oestradiol, ultrasonographic total uterine/individual fibroid volume measurements, and the resistance index (RI) from blood flow velocity waveforms in both uterine and in major fibroid arteries. Significant hypo-oestrogenism was sustained in all patients throughout treatment. There were significant reductions in the mean total uterine (42.1%, SE 2.5) and individual fibroid (59.6%, SE 5.6) volumes at the end of treatment. Significant increases were observed in the mean RI values in both the uterine ($P<0.001$) and the major fibroid ($P<0.001$) arteries, signifying marked reduction in flow within these vessels. This suggests that the reduction in volume may be related to hypo-oestrogenic mediated reduction in flow, and that these agents may be beneficial if given immediately before surgery for fibroids.

Doppler examination of the fetal cerebral circulation in labour by A. Dougall, G. D. Lang & D. H. Evans*. Departments of Obstetrics and Gynaecology and *Medical Physics, Leicester Royal Infirmary, Leicester

Intrapartum asphyxia can result in cerebral damage with short- and long-term sequelae. In the fetal lamb there is redistribution of blood to the cerebral circulation in acute hypoxia. The advent of Doppler ultrasound has permitted the development of non-invasive techniques to study the human fetal circulation. We have developed a method for studying the flow velocity waveforms in the anterior cerebral arteries of human fetuses during labour. Using continuous wave Doppler (Sonicaid Vasoflo 2, 8

Mhz probe), signals were obtained from the anterior cerebral arteries of 43 fetuses. The mean values of Pourcelot's resistance index (PI) were 0.66 (range 0.53–0.97) in early labour, 0.66 (range 0.48–1.00) in late labour and 0.73 (range 0.61–0.78) in the second stage of labour. In 13 patients recordings were made during and between contractions. The mean PI during contractions was 0.89 compared with 0.68 between contractions ($P<0.01$ Wilcoxon one-sample test); the difference in heart rates was not statistically significant. We could not identify any trend in the mean velocities; this is probably a reflection of the alterations in the angle of insonation as a result of movement of the fetal head. This new transvaginal technique offers exciting possibilities in studying the physiology of fetal cerebral blood flow in labour and in the detection of intrapartum hypoxia.

A review of 179 cases of fetal-maternal macrotransfusion by T. C. Li & D. R. Bromham. University Department of Obstetrics and Gynaecology, St James University Hospital, Leeds

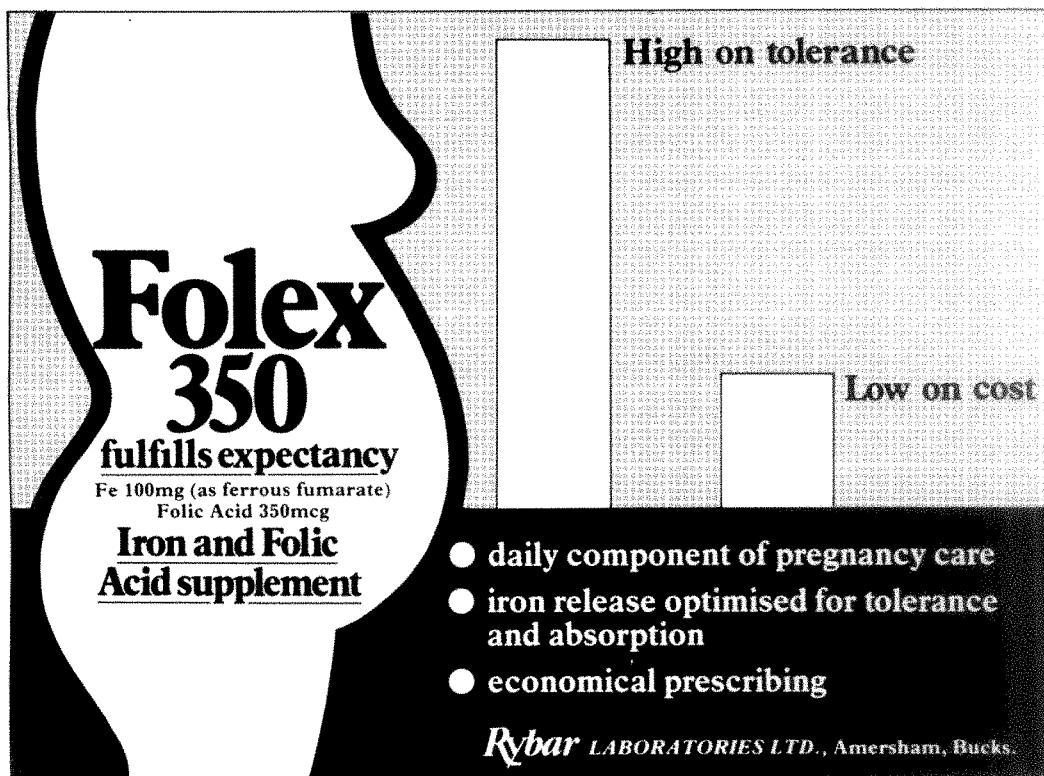
The Kleihauer method to estimate the size of fetomaternal bleeds in a rhesus-negative patient following delivery has been standard obstetric practice in this country for many years. We reviewed the results available over the last 9 years from 11 hospitals in the Yorkshire Region, and identified 179 cases where the volume of bleed was estimated to be 10 ml or more (fetomaternal macrotransfusion, FMMT) and compared these with 322 control cases selected from the same population. Obstetric and perinatal factors in the two groups were compared. Fetal distress in labour was 3.6 times as common and manual removal of the placenta 12 times more frequent in the study group ($P<0.0001$ in both cases). Low Apgar scores (≤ 4) at 1 min were 3.5 times more frequent in the study group ($P<0.05$). The cord haemoglobin value of the study group (15.8 (SE 0.3) g/dl) was significantly lower ($P<0.0005$) than that of the controls (16.7 (SE 0.1) g/dl). There were inverse correlations between the estimated volumes of FMMT and Apgar scores at 1 min ($r=-0.2$, $P<0.0001$) and cord haemoglobin levels ($r=-0.5$, $P<0.0001$). The perinatal mortality and perinatal morbidity in the study group (44 per 1000 and 95 per 1000 respectively) were significantly higher ($P=0.01$ and $P<0.005$ respectively) than those of the con-

trol group (6 per 1000 and 25 per 1000 respectively).

Reticulocytes in the normal puerperium and following postpartum haemorrhage by T. H. Bloomfield. Department of Obstetrics and Gynaecology, Northwick Park Hospital, Harrow, Middlesex

Because blood loss at delivery is difficult to estimate routinely and because haemoglobin concentration may fall during the first few days after delivery as body fluids redistribute, postpartum haemorrhage may be underdiagnosed at delivery and overdiagnosed retrospectively on the ward. Fourteen normal primigravidae were studied. The blood loss at delivery was measured colorimetrically after washing out soiled swabs and linen in water. Venous blood was taken at delivery and daily thereafter for 5 days. Routine haematological indices were measured on a

Coulter model S+IV and reticulocytes counted using a Miller square. Blood loss at delivery ranged from 50 ml to 670 ml, excluding any blood trapped in the maternal surface of the placenta, and there was no change in the reticulocyte count in this group. A second group of 20 normal women were studied weekly for 8 weeks postpartum. In this group, serum erythropoietin and ferritin levels were also estimated by radioimmunoassay. Blood loss at delivery ranged from 17 ml to 1100 ml and the mean haemoglobin rose from 12.4 g/dl before delivery to a maximum of 13.6 g/dl 2 weeks postpartum. Erythropoietin fell from 50 u/l to 25 u/l over the same period. On average the reticulocyte count reached a nadir by 4 weeks postpartum (38 000/ml–20 000/ml) while ferritin levels rose from 30 µg/l at delivery to 77 µg/l at week 8. It is suggested that a fit young mother can tolerate a blood loss of up to a litre at delivery.



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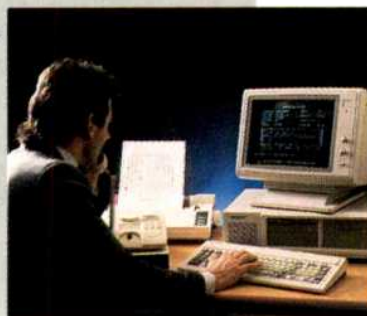
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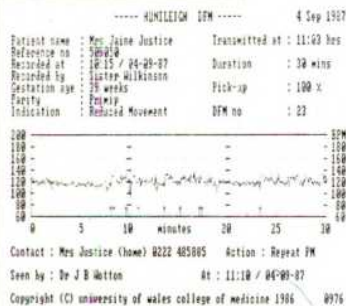
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CORRESPONDENCE

Psychosexual trauma of an abnormal cervical smear

Dear Sir,

We were interested to read the article by Campion *et al.* [*Br J Obstet Gynaecol* (1988) **95**, 175-181], as the findings reported overlap with some of our own, first presented at the annual meeting of the British Society for Colposcopy and Cervical Pathology in June 1985. We studied (funded by the Cancer Research Campaign) the impact of an abnormal cervical smear (and subsequent investigation and treatment) on 153 women in two centres. Our findings and recommendations are shortly to be reported in full by Posner & Vessey (1988). We investigated levels of stress and anxiety, the provision of information and support, the occurrence of symptoms related to diagnostic procedures and treatment, and changes in body image and perception of health status.

We found that some 43% of women reported psychosexual disturbance around the time of investigation and treatment. This disturbance continued in a proportion of women (14%) 6-9 months later. The upset appeared to be related to guilt and resentment about the sexually transmitted nature of the condition, to the negative metaphorical image of cancer which was generally held, and to the unavoidable invasion of 'private space' involved in the medical intervention. Our report describes how this felt from the point of view of the patients, and we hope the information provided will increase understanding of the 'psychosocial' (rather than medical) meaning of this preventive intervention. Questioning about sexual history was felt by many women to be a further intrusion into their privacy and we suggest such questions should be avoided unless they are strictly necessary in clinical or research terms. At the time we conducted our work there was less awareness than now of the male role in the aetiology of cervical neoplasia, and few women expressed hostility towards sexual partners. There was, however, considerable resentment about the stereotypical

media image of the sort of woman most likely to get cervical cancer or its precursors.

In order to avoid unnecessary distress it is essential that women are given adequate information about the nature of cervical intra-epithelial neoplasia (CIN) and its treatment at as early a stage as possible. It is also helpful for the general practitioner or whoever informs the patient of the abnormal smear finding to reassure her that she need not discontinue sexual intercourse. Some women in our sample expressed concern that by continuing they might make the condition worse, or pass it onto their partners, or that they might no longer be acceptable to their partners because they were somehow 'unclean'. Changes in contraceptive practice at this time may add to a woman's anxieties, although the use of a condom should be encouraged. Young women without steady partners appear to be particularly vulnerable to upset in this sphere. We welcome the attention the study of Campion *et al.* has focused on the possible need for sexual counselling.

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Commentary: The safety of diagnostic ultrasound

Dear Sir,

Dr Meire's commentary [*Br J Obstet Gynaecol* (1987) **94**, 1121-1122] on the safety of diagnostic ultrasound has rightly summarized the difficulties in deciding the potential risks of this useful clinical tool. Despite the present chaos in delineating and controlling exposure conditions and the bewildering range of ultrasound bio-effects described in over 700 publications since 1950, reviewed by Stewart & Moore (1984), some order is emerging.

The increased frequency of sister chromatid exchanges induced by pulsed ultrasound in human lymphocytes, first described by Liebeskind *et al.* (1979), has been amply confirmed in reports from four independent laboratories involving studies of pulsed as well as continuous wave ultrasound (Haupt *et al.* 1981; Ehlinger *et al.* 1981; Ozawa *et al.* 1984; Stella *et al.* 1984). Recently, further evidence that sister chromatid exchanges in human lymphocytes are induced by high-intensity pulsed ultrasound has been presented by Barnett *et al.* (1988), who are now able to confirm the previous results.

Free radical production in amniotic fluid and blood plasma by medical ultrasound, probably following gaseous cavitation, has been detected by Crum *et al.* (1987). This provides a likely mechanism for the origin of the DNA damage. Because of these confirmations and a recent report by Ellisman *et al.* (1987) that diagnostic levels of ultrasound may disrupt myelination in neonatal rats, the need for regulation, guidance, and properly controlled clinical studies is clear. Clinicians ought to have some estimate of the level of risks associated with ultrasound, as with any other clinical modality.

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- In-utero intravascular transfusion of the fetus for the management of severe Rhesus isoimmunization — a reappraisal**
- Dear Sir,
- We were interested to read the report by MacKenzie *et al.* [*Br J Obstet Gynaecol* (1987) **94**, 1068-1073] describing the rather disappointing perinatal results obtained with intrauterine intravascular transfusion in the treatment of severe Rh-haemolytic disease.
- In our country, we still have a rather high incidence of severe fetal erythroblastosis due to flaws in our prophylactic plans. Encouraged by the satisfactory results reported by Rodeck *et al.* (1981, 1984) among others (see reference list), we have performed 90 ultrasound-guided intravascular transfusions on 33 patients (1-8 pro-

cedures per patient) in the last 18 months. Mean gestational age at the initial transfusion was 25 weeks (SD 3; range 20–31). Of the 33 patients 64% had a history of intrauterine fetal death and 16 (48%) of the treated fetuses were hydropic at the initial transfusion (ascites, oedema and pericardial effusion). Of the 33 fetuses 12 (36%) died *in utero*, and of the 16 hydropic fetuses 4 (25%) showed reversal of their hydropic condition. Twenty-one (64%) treated fetuses were born alive; of these, 11 died in the early neonatal period (4 of hydrops fetalis, 1 of cardiac arrest during the exchange transfusion, 2 of cardiac failure, 3 of hyaline membrane disease, and 1 of extreme immaturity). Our final neonatal survival rate with the use of intrauterine intravascular transfusions was similar to that we obtained with intrauterine intraperitoneal transfusions.

Our results using intravascular transfusions are very disappointing, as are those obtained by MacKenzie *et al.* (1987). At present, we are trying to find the reasons for these unfavourable results. Our analysis of anti-D levels ($\mu\text{g/ml}$) before and after intravascular transfusion revealed a surprisingly marked rise: in one case anti-D titres increased from 7.31 to 35.2 $\mu\text{g/ml}$. We wonder whether in these severely affected fetuses the vascular disruption caused by the needle during repeated intravascular transfusions enhances anti-D antibody production due to feto-maternal haemorrhage.

We believe that the frequency of intravascular transfusions of the fetus, as well as the timing of the initial transfusion should be reconsidered in order to obtain a better perinatal outcome. Also, plasmapheresis, associated with intrauterine intravascular transfusions could be useful to lower anti-D titres.

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Postpartum haemorrhage—a continuing problem

Dear Sir,

We wish to support the report by Gilbert *et al.* [*Br J Obstet Gynaecol* (1987) **94**, 67–71] in concluding that postpartum haemorrhage (PPH) is re-emerging as a significant problem.

In reviewing the literature of the last 20 years, we found very few reports on PPH; furthermore the average bleeding during the third stage of labour and the following 2 h varied between authors according to the different methods used to quantify the bleeding. In addition, we noticed that some authors did not explain how they had measured the blood loss. However, they all agreed on the definition of PPH, conventionally defined as a blood loss >500 ml after the birth of the baby; PPH occurred in about 5% of all deliveries and induced labours seemed to have a higher risk than spontaneous labours.

Table 1. Postpartum haemorrhage (PPH) in Monza (1984-1985)

	Primigravidae		Multigravidae		Total <i>n</i>
	Total <i>n</i>	No. with PPH <i>n</i> (%)	Total <i>n</i>	No. with PPH <i>n</i> (%)	
Spontaneous labour	1606	336 (20.9)	1300	206 (15.8)	2906
Induced labour	84	21 (25.0)	140	29 (20.7)	224
Total	1690	357 (21.1)*	1440	235 (16.3)	3120

*Significant difference between the two parity groups $P < 0.001$.

We analysed the computerized data on 3731 deliveries in our Institution between January 1984 and December 1985: 3130 were vaginal deliveries of which 2906 were spontaneous and 224 induced (7.2%). In our Institution, either to augment or to induce labour, we use low amniotomy supplemented by intravenous oxytocin according to the protocol described by O'Driscoll & Meagher (1980). As a rule, we did not follow an active mechanical assistance during the third stage of labour, but an intramuscular injection of ergometrine (0.4 mg) was always given after the delivery of the placenta. The blood loss was measured with a plastic graduated sac.

Our data, summarized in Table 1, show that the rate of PPH (18.9%) was higher than the values reported in the literature. The rate of PPH was higher in induced than in spontaneous labours but this difference was not statistically significant. However, as reported by Gilbert *et al.* (1987), PPH was significantly more common among primigravidae. On the other hand, in our sample population the frequency of high parity (≥ 4) was only 0.9%. There are three possible explanations for these findings: first, accurate measurements of blood loss; second, wrong

timing of oxytocin administration [Sorbe (1978) found that oxytocin administration at delivery of the anterior shoulder reduced the frequency of PPH by about 10%]; third, it is possible that the high dosage of intravenous oxytocic used in our sample population was greater than necessary.

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Italy

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BOOK REVIEWS

A Manual of Labour Ward Practice by Malcolm Pearce & Shirley A. Steel. John Wiley & Sons, Chichester, 1988, 283 pages with figures and tables, £17.50

I thoroughly enjoyed reading this book which represents an extremely bold, and to my mind creditable, attempt to encapsulate 'state of the art' labour ward obstetrics in one text, a difficult exercise because of rapid changes in obstetric fashion.

As it is based on the labour ward protocols used at St George's Hospital, the text is bound to have a parochial flavour, and clinical methods applicable to a central London teaching hospital may not stand transplantation to the countryside.

Chapters 1-6 include normal labour and delivery. Generally they are well illustrated and workmanlike. Some parts, however, could do with amplification. For instance, both second and third stages of labour combined occupy only two and a half pages of text, which seems to me rather inadequate although one must accept that labour complications are adequately dealt with later on.

The sections on preterm labour and premature rupture of the membranes are very good, and the chapter on pre-eclampsia and eclampsia does contain some discussion of the use of magnesium sulphate which will please our transatlantic friends. There are a few omissions worthy of note, for instance in the postpartum treatment of severe pre-eclampsia and eclampsia there is no mention of the need to provide adequate analgesia.

Some would take issue with the section on acute pulmonary oedema where physical examination of the cardiovascular system takes a back seat in favour of blood gases and X-rays. The use of colloid solutions in the management of this condition is controversial and potentially dangerous without a Swan-Ganz catheter.

Chapter 14 describes coagulopathies as they affect the obstetric patient. There are nice diagrams of the clotting systems and good advice,

the most important item of which is that a haematologist should first be consulted.

Fetal distress is well dealt with, with good illustrative CTGs and a very good chapter on maternal collapse with subsections on septicæmic shock, amniotic fluid, embolism and pulmonary embolism and pulmonary aspiration syndrome giving good guidance and useful checklists.

There are clear well-illustrated chapters on breech delivery, abnormal cephalic presentation, cord prolapse and uterine inversion.

A useful selection on how to treat various maternal injuries is competently dealt with but the statement that colporrhæxis—rupture of the vaginal vault—'is usually due to . . .', could make the untutored reader feel that this ghastly accident is commonplace. Basic instruction is given in the application and use of forceps. Simpson's long forceps are called 'straight forceps' throughout the chapter, a misnomer—the term 'straight' should rightly be reserved for forceps without a pelvic curve, if memory serves correctly!

The short (three and a half page) chapter on the obstetric flying squad is inadequate for units where flying squad calls are frequent. The recommended set of equipment fails to include drugs to treat eclampsia, and no equipment for the resuscitation of the convulsing or unconscious mother is provided—a serious omission. The text is very up to date in certain areas and in a sense reflects the problems of the changing world we live in; useful chapters on the management and labour of patients with haemoglobinopathy, Australia-antigen positive patients and those afflicted with AIDS are very well done.

The advice given for the management of still-birth and neonatal death is excellent and a useful checklist is provided.

There is also a good chapter on 'legal aspects', with some discussion of 'birth plans'. These are still somewhat of a novelty for some of us. The advice given to readers regarding the views of individual women, including problems posed by

Jehovah's witnesses, seems to be sound and based on practical experience.

At the end there is a glossary of over 100 abbreviations many of which throw new light on labour ward 'newspeak'. For instance we are informed that AA now stands for 'Australia-antigen positive patient' (not Alcoholics Anonymous) and more germanely POP stands not for 'persistent occiput posterior' but 'progesterone only pill', and so on.

As obstetric fashions change it is incumbent upon all involved in childbirth continually to update their labour ward procedures. I think Mr Pearce and Dr Steel have done most of us a service in that the contents of this book can be used as a series of procedural models around which we can draw procedures applicable to our local needs, and would also provoke lively debate.

James F. Pearson

Fertility and Family Planning in the Third World: A Case Study of Papua New Guinea edited by William K. A. Agyei. Croom Helm Ltd, Beckenham, 1987, 208 pages, £19.95

This is an account of a demographic survey carried out by the author in Papua New Guinea in 1979/80. His aim was to provide information on fertility, infant and child mortality, and contraceptive practices among the population which would provide the basis for the formulation of a population policy.

Although the book is unlikely to be of interest to the practising gynaecologist in the UK, it provides useful background information for doctors whose work involves family planning programmes in developing countries and, of course, in Papua New Guinea in particular.

After an introductory chapter on demographic changes and family planning practices in the Third World, the author recounts the methods whereby he obtained information from 6391 adult men and women from the four geographical areas of Papua New Guinea. In a country of high fertility, with a population growth of nearly 3% per annum, large families are the norm, and even in urban areas the ideal family size is over three. Although infant mortality rates have declined considerably over the last 15 years, they still range between 80 and 100:1000. Most babies are breast-fed for nearly 2 years and many couples practise sexual abstinence during this time 'to ensure the welfare of

the mother and the child'. The Papua New Guinean government has promoted breast-feeding and a doctor's prescription has to be obtained for artificial milk feeds. However, very few had any knowledge of the inhibitory effect of breast-feeding on fertility.

Between a quarter and a half of those questioned were aware of the common methods of family planning, but very few used them, and there was a general attitude that modern contraception was a danger to the health of both mother and offspring. Although contraception is only available to women in Papua New Guinea with their husband's consent and only 13 had been sterilized, very few men used contraception, and not one of those questioned had had a vasectomy.

Because I have worked in Papua New Guinea, I found this book quite enjoyable, but I would not recommend it to anyone who does not have a particular interest in that country, or in Third World attitudes to family planning.

Jill Everett

Caesareans: An Explanation and Preparation by Elliot Philipp. Sidgwick & Jackson, London, 1988, 140 pages with figures and tables, £12.95

With caesarean section rates now approaching 20% in many NHS delivery units, and even higher rates in private practice, Elliot Philipp and his publishers have clearly anticipated a growing need for a layperson's text explaining the operation. However, he goes much further than simply describing the surgical procedure and its indications and complications. He has discussed at length the medical and legal background which has caused the proportion of abdominal deliveries to quadruple in the last 30 years. He treats the issues with commendable candour and realism, for example devoting seven pages to why caesareans are sometimes performed at the mother's request, for the doctor's convenience (especially in private practice), for insurance purposes, and because of medico-legal considerations. Less mundane aspects are also covered, such as malposition, caesarean hysterectomy and the psychological consequences of the operation.

The text is written in Mr Philipp's inimitable style, which is chatty without being longwinded. There is a fair sprinkling of instructive stories which help to maintain interest. Most of the

statements in the book seem to be a fair consensus of current opinion, which is not surprising given the 30 or so people listed in the acknowledgments. There are one or two odd mistakes, such as the suggestion on page 43 that ergometrine was discovered and introduced in the 1940s (in fact it was widely used, as extract of ergot, in the last century, and purified by J. H. Burn in the 1920s). Even more curious is the statement on page 134 that 'the trend for doing more and more caesarean sections started after the discovery of antibiotics in the 1960s'. Working as I do at St Mary's in Paddington, I can hardly believe that anyone is unaware that the first antibiotic (Salvarsan) was used in the UK by Alexander Fleming in 1910 (and reported in the *Lancet* in 1911) and that Fleming himself discovered penicillin at St Mary's in 1928. However, such odd lapses are fortunately not typical.

As a professional rather than a consumer, it is not possible for me to tell whether Mr Philipp has judged his market exactly. However, I enjoyed reading the text myself and would certainly recommend it to my more literate and questioning clients. Only time, and sales figures, will tell whether this book will be a hit—but I suspect it will.

P. J. Steer

Neonatal and Perinatal Medicine: Diseases of the Fetus and Infant. Fourth edition edited by A. A. Fanaroff & R. J. Martin. The C. V. Mosby Company, St Louis, 1987, 1232 pages with figures and tables, £75.00

Neonatologists have no shortage of textbooks claiming to cover the whole of their field, but one wonders whether the specialty has progressed to the point where it is no longer feasible to be comprehensive, at least at the postgraduate level. This 3.5-kg baby is an impressive all-American rewrite of Behrman's *Neonatal-Perinatal Medicine*, and its 40 chapters and three appendices do just about cover the lot. Like many American texts it is well illustrated, well laid out and easy on the eye. There is a good balance between clinical medicine, basic science and practical advice. The index is excellent and very comprehensive. A personal dislike, however, is the use of bibliographies at the end of chapters instead of numbered references. While 'further reading' can be useful for students, and leaves the chapters looking less cluttered, it is

not possible for the more advanced reader to tell whether a particular point made by an author is purely didactic or referable to source. New chapters include those on regional planning, staff psychology (studied more often than infant psychology these days) and bioethics. Although the latter naturally refers to certain legal points applicable to the US, it is well written and of wider interest.

The antenatal aspects of perinatal care are given rather wider coverage than in many neonatal texts, but there are odd lacunae such as the management of antepartum haemorrhage and its relationship to outcome. Recent 'hot topics' such as the evolution and diagnosis of periventricular leucomalacia, retinopathy of prematurity and AIDS, get surprisingly short, albeit accurate, coverage. Relatively little space is given to the use of diagnostic ultrasound postnatally except in the cardiology section, where a few of the scans are reproduced very poorly. Cranial ultrasound is barely mentioned and not really illustrated, and hydrocephaly is demonstrated by air-encephalogram.

The British chauvinist neonatologist would comment on the paucity of references to UK work, and that he had just bought Robertson's *Textbook of Neonatology* (same weight and price). There is perhaps little to choose between them, and both provide a solid starting point for the neonatal reader. This book is perhaps stronger on fetal aspects, but the jokes are better in 'Robertson'.

Richard Cooke

Haines and Taylor Obstetrical and Gynaecological Pathology. Third edition edited by H. Fox. Churchill Livingstone, Edinburgh, 1987, Vol. 1: pp. 1–696; Vol. 2: pp. 697–1331 with figures and tables, £195.00

Haines and Taylor has changed out of all recognition. The new third edition comes in two volumes of larger format with a resultant weight gain of more than 4 kg. Although the names of the two original authors have been retained as a tribute to two pathologists, both now deceased, whom many consider to be the fathers of modern British gynaecological pathology, the text has been completely rewritten. In this context, it might have been preferable if all the authors had been British but instead, the expertise of 56 authors from several disciplines and many dif-

ferent countries has been used. They have been allowed to write in their individual styles and the variety is usually refreshing rather than distracting. Some clinicians have been included, and this, together with good clinical correlation throughout should make the book of value to practising gynaecologists as well as to pathologists. There is even a joint attack on the nomenclature of vulval disorders by a dermatologist and a pathologist which helps to dispel much long-standing confusion.

Volume 1 deals with gynaecological subjects, covered in the traditional order from vulva to ovary (though it is unfortunate that ovarian tumours overflow into the other volume). There are also excellent introductory sections on embryology, anatomy and abnormal sexual development.

Volume 2 deals mainly with the pathology of pregnancy and related topics (including infertility, abortion, perinatal pathology, maternal mortality and trophoblastic disease) and is especially valuable as this information is not readily found elsewhere.

The text is liberally illustrated, mainly by photomicrographs. These are well chosen and usually of good quality though some are too dark and others lack the clarity of reproduction seen in the earlier editions, even when the same photographs are used.

Each chapter is followed by a long list of useful references though few are more recent than 1984.

Typographical errors are very few (although the legends to figures 39-20 and 39-21 appear to have been transposed).

This edition, like the first, is a landmark in gynaecological and obstetrical pathology and the wealth of information it provides should be available to all gynaecologists and their pathologist colleagues though the price might be a deterrent.

A. S. Hill

Books received some of which may be reviewed at a later date

Parasitic Infections in Pregnancy and the New-born edited by Caroline MacLeod. Oxford University Press, Oxford, 1988, 308 pages with figures and tables, £35.00

Current Therapy of Infertility (3) edited by Celso-Ramon Garcia, Luigi Mastroianni Jr, Richard D. Amelar & Lawrence Dubin. B. C. Decker, Inc., Toronto, Philadelphia, 1988, 334 pages with figures and tables, £36.50

Progress in Infertility Third edition edited by S. J. Behrman, Robert W. Kistner & Grant W. Patton Jr. Little, Brown & Company, Boston/Toronto, 1988, 852 pages with figures and tables, £65.00

Atlas of Hysterosalpingography by Stephen Karasick & David Karasick. Charles C. Thomas, Springfield, Illinois, 1988, 192 pages illustrated, \$46.75

Pathology of Infertility: Clinical Correlations in the Male and Female edited by Bernard Gondos & Daniel H. Riddick. Thieme Medical Publishers, Inc., New York, 1987, 388 pages with figures and tables, DM 189

The Physiology of Reproduction Volumes I and II edited by Ernst Knobil & Jimmy D. Neill. Raven Press, New York, 1987, 2633 pages, \$362.50

Coroners' Records in England and Wales edited by Jeremy Gibson & Colin Rogers. Federation of Family History Societies, 1988. Available from J. W. S. Gibson, Harts Cottage, Hanborough, Oxford OX7 2AB or The Federation of Family History Societies, c/o Benson Room, Birmingham and Midland Institute, Margaret Street, Birmingham B3 3BS. £1.50 (+25p postage)

Baillière's Clinical Obstetrics and Gynaecology International Practice and Research Vol. 1 No. 4. Renal Disease in Pregnancy edited by Marshal D. Lindheimer & John M. Davison. Baillière Tindall, London, 1987, £15.00

COMMENTARY

Broken bones—a gynaecological problem

Most gynaecologists might well react with suspicion and disbelief if it were put to them that there exists a preventable condition, effectively untouched by our speciality though largely caused by ovarian dysfunction and which costs British women untold suffering, the National Health Service £700m per annum and more deaths than cancer of the cervix and ovary combined. Yet such a condition does exist.

Opening a recent meeting organized by the National Osteoporosis Society, Edwina Currie, MP, voiced some welcome ministerial concern over the fact that in 1985, 35 000 women sustained femoral neck fractures in England and Wales, an incidence which is showing a true increase after allowance for the demographic expansion of the population at risk (Boyce & Vessey 1985). The bald figure, however, fails to transmit the scale of mortality and morbidity which osteoporosis-related fractures bring in their train. Of every 100 women who sustain a femoral neck fracture up to 20 will die as a result and 50 will not return from hospital to their own homes (Melton & Riggs 1983). Osteoporosis is thus not only costing women their lives but also robbing them of what is often their most cherished social attribute—their independence.

Hitherto, the attitude of the profession towards prevention of this condition has been somewhat nihilistic. Osteoporosis, it has been said, is not a true bone disease but simply a progressive diminution in the amount of normal bone. Promoted as it is by the physiological menopause, it is an inevitable accompaniment of ageing and is thus part of the natural order. *Ergo*, it is to be accepted and borne with equanimity. Now this smacks of the Old Testament view of labour: 'in sorrow thou shalt bring forth children' (Genesis 3, 16). As obstetricians, we and our anaesthetic colleagues have effectively seen off this scriptural passivity. As gynaecolo-

gists, should we attempt the same in the case of osteoporosis?

Part of the problem with this condition is that it is something of a clinical orphan, with foster parents ranging across the disciplines of endocrinology, metabolism, orthopaedics and gynaecology. While the management of the established clinical disease will properly remain the concern of our colleagues in general medicine and orthopaedics, what is urgently required is a dedicated adoptive parent to take on the critical area of prevention. Given that the disease is largely one of the female sex, that the prime cause of accelerated bone loss is climacteric ovarian failure, that in hormone replacement therapy we now have regimens which appear both effective and safe in the long term—given all that, it is surely the gynaecologist and the general practitioner who should assume this role.

The natural history of perimenopausal bone loss has been clarified in recent years, together with those factors which influence the two central variables of peak bone mass at maturity and rate of subsequent loss in the trabecular bones of spine, hip and forearm. Put simply, the more bone there is to start with and the more slowly it is lost, the less likely is an individual to reduce her bone mineral content (BMC) to a level where fracture is a threat. Thus black women with their high bone mass at maturity do not in general suffer from osteoporosis although they experience the menopause and subsequent bone loss just as do white women. Conversely the patient most likely to develop osteoporosis is a fair-skinned, asthenic, sedentary white woman with an early menopause and a penchant for tobacco or alcohol. To date no universal test for bone mineral content has found favour and the absorptiometric bone scan of the forearm, though easy to perform, has failed to live up to its early billing as the pap smear of the skeleton.

Recently, however, it has been claimed that a function derived from a single measurement of height (for fat mass) together with serum alkaline phosphatase, urinary calcium and hydroxyproline will correctly identify 80% of those patients with the highest bone loss rates (Christiansen *et al.* 1987). These data await confirmation.

Even without screening, so general is age-related bone loss that it is possible to estimate bone mineral content to within 10% using age alone. Women can expect to lose 50% of their mature trabecular bone mass in spine, distal radius and femoral neck during an average lifespan as well as 35% of their cortical bone (Riggs 1987). It appears that there is a steady background attrition, though at different rates, in trabecular and cortical BMC from about age 40 in both sexes. In the case of women, however, there is superimposed upon this steady loss approximately a decade of accelerated bone loss due to the perimenopausal decline in oestrogen production. Thereafter rates of loss decline and become asymptotic with the background rate. In practical terms then, it is during the critical postmenopausal decade that preventive action is required. What can be done?

The three main areas in which the doctor can offer advice to the woman worried about osteoporosis are exercise, diet and hormone replacement therapy (HRT). There is no doubt that the pull of muscle upon bone is essential for the health of the latter and women should be encouraged to have an active programme of exercise. However, neither exercise alone nor combined with a diet containing adequate calcium is sufficient to protect the BMC and it does seem that an additional specific measure is required to inhibit the excessive bone resorption. A review of all the agents claimed to prevent BMC loss is beyond the scope of this commentary. However, it has been repeatedly and clearly shown that oestrogen is capable of inhibiting bone resorption although its precise mechanism of action awaits final clarification. Despite a determined hunt, oestrogen receptors have yet to be conclusively identified in bone tissue. Nevertheless, an increasing volume of data suggests that in appropriate dosage—such as 0.625 mg conjugated oestrogen or 2 mg oestradiol orally per day or equivalent by implant or transdermal patch—oestrogen works safely and with acceptability to the patient.

A gynaecologist faced with requests from

healthy patients for HRT will have to weigh considerations which will be both theoretical and severely practical. A prime example of the former will certainly be—*Primum non nocere*—am I likely to occasion short- or long-term harm to this patient by prescribing HRT? An example of the latter might be—what on earth will happen to my outpatient clinic if I embark on wholesale HRT with regular if infrequent review? On the first point reassurance is indicated. Given a good history and clinical examination to uncover the few absolute and main relative contraindications, our colleagues can be assured that HRT is unlikely to harm the patient. Indeed on the major issue of cardiovascular disease the evidence generally points to a protective effect, particularly with regard to coronary artery disease (Ross *et al.* 1981). Oestrogen without question protects the skeleton against excessive bone resorption and the addition of a progestogen to induce either regular endometrial shedding or amenorrhoea now has the added bonus that certain of these agents appear to inhibit bone loss in their own right (Selby *et al.* 1985).

On the practical side there is much to be said for minimizing the effects of HRT on the outpatient clinic by organizing a regular menopause clinic run either by a consultant with an interest, a junior in training or, as in many successful cases, by a clinical assistant. Such a specialist clinic frees the general clinic from potential overload, provides excellent clinical and educational benefit for juniors—and for medical students if present—and encourages research through availability of numbers.

With regard to perhaps the most vexed question of all—how long to treat—the definitive answer is still awaited. There will always be those who view the menopause as analogous to adrenocortical failure or hypothyroidism, a true endocrine deficiency state requiring treatment for life. Quite apart from ultimate patient non-compliance with this approach, it is inherent in gynaecologists to be cautious in the use of powerful hormones with multisystem effects and the practical expectation is that, given clinical reassurance on safety, a finite period of HRT is probably the desired goal. But precisely how long? With regard to osteoporosis, light at the end of this particular tunnel is perhaps perceptible through the mathematical modelling work of the MRC Bone Mineralisation Group at Leeds (Horsman & Marshall 1986). Using a stochastic

approach which allows the incorporation of random incidents such as falls, the likelihood of osteoporosis-related fractures in elderly female groups can be calculated. Starting from the basic principle that BMC at any age and site is determined by absolute amount of bone at maturity and to rate of postmenopausal loss, it is possible to predict not only the likelihood of fracture but also the impact of any regimen, such as HRT, which modifies rates of BMC loss at specific sites.

Using this model, it can be predicted that in an unselected UK white population, the abrogation of postmenopausal bone loss by HRT for 10 years would result in a 40–45% reduction in the incidence of femoral neck fracture, while the treatment of a selected 'low quartile' (that 25% of women with least bone at menopause) would reduce the total incidence of fracture by the same amount. Here then is a hypothesis that by a finite decade of prophylaxis from, say, age 50–60 years, BMC can be afforded such protection that even with resumed bone loss thereafter, women may reach their present expectation of fourscore and two with sufficient bone mineral to withstand significant impact loading without fracture. In other words, they may achieve the desired end points of regaining their feet intact after a fall and of finding their stature undiminished with the passing of the years.

Studies underway in the UK, the rest of Europe, and the USA are likely to shed more light on optimal dosage, route and duration of HRT regimens in the near future. For the present, the door stands open for the prevention of one of the major public health problems of modern times. By the year 2000 the number of women aged 65 and over will increase by half

(Riggs 1987). The woman who will then be 65 is already 53, had her menopause 2–3 years ago and, probably untreated, is losing bone. It is surely for us as gynaecologists to seize the initiative, go out to our communities together with our partners in primary care and take on board the active prevention of this fearsome condition. To do so would simply be to keep faith with the central objective of our speciality, the pursuit of excellence in the care of women.

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Ectopic pregnancy in Aberdeen 1950-1985

G. M. M. FLETT, D. R. URQUHART, C. FRASER, P. B. TERRY,
J. C. FLEMING

Summary. The reported incidence of ectopic pregnancy in Aberdeen City and suburbs (1950-1985), using as denominators maternities, pregnancies and women aged 15-44 years, has increased threefold since 1970 to 6.4/1000 pregnancies. This increased incidence persisted after the exclusion of previously sterilized women. A total of 11 128 women were sterilized in Aberdeen City and suburbs between 1960 and 1982; 36 ectopic pregnancies occurred in this sterilized population. The prevalence of ectopic pregnancy was 3.55/1000 sterilizations. This did not alter significantly over the period of study despite changes in the method of sterilization. However, due to the increased popularity of sterilization, the proportion of ectopic pregnancies in women who had been sterilized increased from 0% in the 1950s to 21% in the quinquennium 1975-1979.

Ectopic pregnancy remains a serious acute gynaecological emergency and still contributes to maternal mortality (DHSS 1985). The incidence of ectopic pregnancy has risen in recent years (Beral 1975; Weström *et al.* 1981; Weinstein *et al.* 1983; Makinen 1987) but the use of different denominators has made comparisons difficult (Barnes *et al.* 1984; Weckstein 1985).

All denominators are unsatisfactory as a measure of women at risk of ectopic pregnancy. The number of women aged 15-44 years may appear to be the most satisfactory, but this takes no account of either changing contraceptive practice or the variation of contraceptive use within the population. The number of maternities is influenced by the increasing incidence of therapeutic termination of pregnancy. Recorded pregnancies in a population probably represents the most accurate measure of women

at risk despite the fact that many early fetal losses are unrecognized or unrecorded.

One aim of this study was to document the changing incidence of ectopic pregnancy and any variation in surgical practice in a geographically defined population over the period 1950-1985.

The reasons for the rising incidence of ectopic pregnancies are uncertain, but several factors have been implicated. Improved diagnostic techniques, resulting in intervention in some cases that might otherwise have resolved spontaneously, and changes in contraceptive practice have been blamed (Vessey *et al.* 1974), as has tubal damage following pelvic sepsis (Weström 1975; Clark & Baranyai 1987) and tubal surgery. Other implicated factors include in-vitro fertilization, postcoital oestrogen contraception, diethyl stilboestrol exposure *in utero* and genital tuberculosis. Abnormal chromosomal patterns occur more commonly in ectopic gestation (Stratford 1970) and while this may explain why ectopic pregnancies occur in tubes without anatomical abnormality, there is no reason to suspect that the incidence of such abnormalities has increased recently.

Failed female sterilization is another factor (Hughes 1980). In a review of 32 papers (McCausland 1980), it was possible to examine the sterilization failures in terms of intrauterine

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and ectopic pregnancies: 12.3% of pregnancies after non-laparoscopic tubal sterilization and 51% of those after laparoscopic tubal coagulation were ectopic. In this comparison between non-laparoscopic sterilization and laparoscopic tubal coagulation, the calculated prevalence of ectopic pregnancy in the population of sterilized women was approximately 1/1000 in both groups. The range varied from 0 to 21.9/1000 sterilized women. For clip sterilization, 4.4% of failures were reported as ectopic pregnancies (Tatum & Schmidt 1977) again giving a calculated prevalence of ectopics of 1/1000 sterilizations.

In a study at Dulwich Hospital between 1970 and 1973 (Chakravarti & Shardlow 1975) tubal sterilization had preceded 12% of a consecutive series of 50 ectopic pregnancies. To balance this risk, however, the likelihood of an ectopic pregnancy following tubal sterilization has been estimated to be lower than in the absence of sterilization or other contraceptive practice (De Stefano *et al.* 1982). The long-term follow-up of large numbers of patients is difficult due to patient mobility, but the Aberdeen area has the advantage of a stable population served by one large teaching hospital. Most studies of ectopic pregnancies after sterilization have involved small numbers and have not expressed the incidence of ectopic pregnancy after sterilization in terms of the number of women sterilized or the sterilized population of childbearing age. Another aim of this study was to examine the occurrence of ectopic pregnancy after sterilization in terms of the total number of ectopic pregnancies, the number of sterilizations undertaken and the sterilized population of childbearing age in a well-defined geographical area, taking account of changing modes of sterilization as far as possible.

Patients and methods

Recorded information for all patients in Aberdeen City and suburbs who had an ectopic pregnancy between 1950 and 1985 was obtained from the Aberdeen Maternity Hospital (AMH) data bank. The information obtained included details of the side affected, the surgical procedure performed, the number of pregnancies and the number of maternities. The population of women aged 15–44 years during the study period was obtained from census data (Registrar General, Scotland 1950–1985). Prior to 1974,

boundary changes and less detailed statistics from the Registrar General made the calculation of the female population aged between 15 and 44 difficult. However, an estimate was made using local population data, assuming that the proportion of women in the population aged between 15 and 44 has been the same throughout the study period. In view of the increasing popularity of female sterilization and its known association with ectopic pregnancy, all those women who were previously sterilized were identified from the hospital records and the incidence of ectopic pregnancy in the non-sterilized population calculated. Consideration was given to adjusting the denominators to account for a past history of sterilization. Assuming that 12.3% of the pregnancies after non-laparoscopic sterilization and 51% of those occurring after laparoscopic sterilization are ectopic (McCausland 1980), the number of pregnancies and maternities after sterilization is small (98 pregnancies in the non-laparoscopic sterilized population and 45 pregnancies in the laparoscopic sterilized population). However, the same assumption may not be true for previously sterilized women aged 15–44 and this denominator was reduced by the calculated number of sterilized women aged 15–44 for each quinquennium.

Information relating to 11 128 sterilizations between 1950 and 1982 was obtained from the Aberdeen Maternity Hospital (AMH) data bank. The case records of parous patients with an ectopic pregnancy were examined and the possibility of a sterilization procedure having been undertaken in another centre was excluded. Where ectopic pregnancy was subsequent to a sterilization, details of the sterilization procedure were also obtained from the patients' case records. In earlier years the method of sterilization was not recorded routinely in the database, but examination of a random sample of the records demonstrated that the majority had undergone laparotomy and tubal ligation. From the mid 1970s to 1982, the vast majority of laparoscopic sterilizations were by tubal diathermy. The sterilized female population aged 15–44 years was calculated for each year and the number of sterilized woman-years calculated. Women over 44 years of age were excluded.

Results

The incidence of ectopic pregnancy remained relatively constant in the period 1950–1974 rang-

ing from 2.40/1000 to 3.10/1000 pregnancies (2.87/1000 to 3.40/1000 maternities and 0.20/1000 to 0.27/1000 women aged 15–44) (Table 1). Thereafter the incidence rose to 6.40/1000 pregnancies (8.70/1000 maternities and 0.45/1000 women aged 15–44) in the period 1980–1984, and similar results were obtained after the exclusion of previously sterilized women (Table 2).

In all periods of the study salpingectomy alone was the most commonly performed procedure, but in the earlier years removal of the ipsilateral ovary was undertaken in 33% of cases (Table 3). Recently ovarian conservation has been practised and in the period 1980–1984 fewer than 6% of women with a tubal pregnancy had an oophorectomy. Conservation of the affected tube has become more popular but was still performed in only 7% of patients in the quinquennium 1980–1984 (Table 3).

In the period 1950–1982 inclusive, 413 ectopic

pregnancies were recorded and 36 of these occurred in previously sterilized patients.

During the period 1950–1979 inclusive, there was an increase in the average number of women sterilized from approximately 84/year to 625/year (Table 4). The method of sterilization changed over this period of time such that all women prior to 1965 were sterilized at an open laparotomy but by the quinquennium 1975–1979 over 90% of patients underwent a laparoscopic procedure, predominantly tubal diathermy.

The prevalence of ectopic pregnancy in the sterilized population was around 3.55/1000 and did not change significantly over the period of the study despite changes in the method of sterilization.

Six ectopic pregnancies occurred after 3104 postpartum laparotomy sterilizations, six occurred after 2500 open laparotomy sterilizations, and 23 occurred in the 4261 patients ster-

Table 1. Incidence of ectopic pregnancy in quinquennia (1950–1984) using number of maternities, recorded pregnancies and women aged 15–44 as denominators: Aberdeen City (including ectopic pregnancies after sterilization)

Quin- quennium	No. of ectopic pregnancies	No. of pregnancies	Rate/1000 pregnancies	No. of maternities	Rate/1000 maternities	Average annual no. of women aged 15–44	Average annual rate/1000 women aged 15–44
1950–54	48	18 058	2.66	16 552	2.90	44 800	0.21
1955–59	60	19 371	3.10	17 653	3.40	44 600	0.27
1960–64	51	19 953	2.56	17 765	2.87	43 600	0.23
1965–69	55	19 141	2.87	16 476	3.33	43 300	0.25
1970–74	44	18 359	2.40	14 357	3.06	44 300	0.20
1975–79	89	15 653	5.68	11 469	7.76	45 300	0.39
1980–84	108	16 866	6.40	12 415	8.70	47 600	0.45

Table 2. Incidence of ectopic pregnancy in quinquennia (1950–1984) using number of maternities, recorded pregnancies and women aged 15–44 as denominators: Aberdeen City (excluding ectopic pregnancies after sterilization)

Quin- quennium	No. of ectopic pregnancies	No. of pregnancies	Rate/1000 pregnancies	No. of maternities	Rate/1000 maternities	Average annual no. of women aged 15–44	Average annual rate/1000 women aged 15–44
1950–54	48	18 058	2.66	16 552	2.90	44 716	0.21
1955–59	60	19 371	3.10	17 653	3.40	44 448	0.27
1960–64	50	19 953	2.51	17 765	2.81	43 404	0.23
1965–69	50	19 141	2.61	16 476	3.03	43 039	0.23
1970–74	44	18 359	2.40	14 357	3.06	43 646	0.20
1975–79	70	15 653	4.47	11 469	6.10	44 676	0.31
1980–84	97	16 866	5.74	12 415	7.81	47 000	0.41

Table 3. Ectopic pregnancy, Aberdeen City (1950–1984): affected side and operation

Quinquennium	Affected side			Operation			
	Left <i>n</i> (%)	Right <i>n</i> (%)	Bilateral, unknown or unspecified <i>n</i> (%)	Salping- ectomy <i>n</i> (%)	Salpingo- oophorectomy <i>n</i> (%)	Ectopic removed from fallopian tube <i>n</i> (%)	Unrecorded or others <i>n</i> (%)
1950–54	14 (29.2)	22 (45.8)	12 (25.0)	28 (58.4)	16 (33.3)	0 —	4 (8.3)
1955–59	27 (45.0)	31 (51.8)	2 (3.2)	38 (63.3)	21 (35.0)	0 —	1 (1.7)
1960–64	20 (39.2)	29 (56.9)	2 (3.9)	31 (60.8)	17 (33.3)	2 (3.9)	1 (2.0)
1965–69	26 (47.3)	26 (47.3)	3 (5.4)	38 (69.1)	14 (25.5)	2 (3.6)	1 (1.8)
1970–74	15 (34.1)	29 (65.9)	0 —	35 (79.5)	8 (18.2)	1 (2.3)	0 —
1975–79	37 (41.6)	48 (53.9)	4 (4.5)	76 (85.4)	2 (2.2)	4 (4.5)	7 (7.9)
1980–84	43 (39.8)	59 (54.6)	6 (5.6)	89 (82.4)	5 (4.6)	8 (7.4)	6 (5.6)
Total	182 (40.0)	244 (53.6)	29 (6.4)	335 (73.6)	83 (18.3)	17 (3.7)	20 (4.4)

Table 4. Prevalence of ectopic pregnancy by method of sterilization (1950–1979)

Year of sterilization	Laparoscopic sterilization	Laparotomy	Postpartum sterilization	Total sterilized	No. of ectopic pregnancies after sterilization by quinquennium of sterilization	No. of ectopic pregnancies per 1000 sterilizations for each quinquennium
1950–54	0	320	100	420	0	0
1955–59	0	417	345	762	1	1.31
1960–64	0	396	584	980	4	4.08
1965–69	39	536	730	1305	3	2.30
1970–74	1637	625	1011	3273	15	4.58
1975–79	2585	206	334	3125	12	3.84
Total	4261	2500	3104	9865	35	3.55
Number of ectopic pregnancies after sterilization	23	6	6			
Rate per 1000 sterilizations	5.4	2.4	1.9			

ilized via the laparoscope. Thus, the rate of ectopic pregnancy was 5.4/1000 sterilizations for laparoscopic procedures, 2.4/1000 sterilizations undertaken as open laparotomy, and 1.9/1000 for postpartum sterilization procedures (Table 4). The higher rate of ectopic pregnancy after laparoscopic sterilization was confirmed when the figures were expressed in terms of the number of sterilized woman-years (Table 5). The rate of ectopic pregnancy was 0.70/1000 sterilized woman-years for laparoscopic sterilization, 0.19/1000 sterilized woman-years for laparotomy sterilization and 0.16/1000 sterilized woman-years for postpartum laparotomy sterilization.

When considering the proportion of ectopic pregnancies in women who had been sterilized there was an increase from 0% in the 1950s to 21.4% in the quinquennium 1975–1979 (Table 6). There may be a substantial time interval between sterilization and ectopic pregnancy and in our series the time interval was from 9 months to 19 years but only three ectopic pregnancies occurred more than 7 years after sterilization and 75% occurred within 5 years of sterilization (Table 7).

Discussion

Few studies have examined the incidence of ectopic pregnancies using the three accepted

Table 5. Incidence of ectopic pregnancy per 1000 woman-years by method of sterilization and quinquennium

Year	No. of ectopic pregnancies after sterilization by quinquennium of ectopic	No. of sterilized woman-years (age 15-44 years)	Incidence of ectopic pregnancy/1000 sterilized woman-years
1950-54	0	830	0
1955-59	0	3499	0
1960-64	1	7551	0.13
1965-69	5	12 158	0.41
1970-74	0	20 750	0
1975-79	19	34 308	0.55
1980-82	11	22 617	0.49
Total 1950-1982	36	101 911	0.35
Laparoscopy	24	34 192	0.70
Laparotomy	6	30 760	0.19
Postpartum sterilization	6	36 959	0.16

denominators on the same population, and no other study has been able to take account of the increased incidence of sterilized women. As with all retrospective studies, under-reporting is likely to be the main error, but the AMH data bank represents an accurate source of information on a geographically well-defined population.

The incidence of ectopic pregnancy has risen since 1974 in Aberdeen, the current rate being three times higher than it was in the years prior to the early 1970s. It is possible that such a rise is apparent rather than real. When a patient gives a history suggestive of ectopic pregnancy, a definite diagnosis is now more likely to be made, as a result of laparoscopy, high-resolution ultrasound and sensitive pregnancy tests. In earlier years, conservative management and spontaneous resolution may have occurred (Lund 1955). However, a recent study from Finland

suggests that the observed increase in ectopic pregnancies is only in part due to better diagnosis (Hemminki & Heinonen 1987).

The increased number of ectopic pregnancies after sterilization has been a cause for concern and may account for part of the rise. The study of the relation between sterilization and a subsequent ectopic pregnancy is difficult. The first problem is the time interval that occurs between the sterilization and the ectopic pregnancy. For example, there is still time for some of our women sterilized in the 1970s to develop an ectopic pregnancy. This is one explanation for the apparent fall in the incidence of ectopic pregnancy in patients sterilized in the quinquennium 1975-1979. In the study period, population movement has been immigration rather than emigration. However, it remains possible that a number of ectopic pregnancies have occurred in sterilized patients who have emigrated. In consequence, the true prevalence of ectopic pregnancy is probably slightly higher than reported.

This study has demonstrated that approximately 0.35 ectopic pregnancies occur per 1000 sterilized woman-years. This is far higher than other published studies (McCausland 1980) but it is evident that because of population movements and the length of time between sterilization and a subsequent ectopic pregnancy that under-reporting of ectopic pregnancies in a sterilized cohort is far more likely to occur than over-reporting. It is apparent that an increasing proportion of the women with ectopic pregnancies will give a history of having been sterilized and it is important that clinicians dealing with acute abdominal pain in a previously sterilized woman

Table 6. Proportion of ectopic pregnancies with a history of previous sterilization

Year of ectopic pregnancy	No. of ectopic pregnancies	No. previously sterilized	Per cent of ectopic pregnancies after sterilization
1950-54	48	0	0
1955-59	60	0	0
1960-64	57	1	2
1965-69	55	5	9
1970-74	44	0	0
1975-79	89	19	21
1980-82	66	11	17

Table 7. Interval between sterilization and ectopic pregnancy (1950–1982)

Time interval	No. of sterilized patients developing ectopic pregnancy
<1 year	6
1–3 years	11
3–5 years	10
5–7 years	6
>7 years	3

are not lulled into a false sense of security by the history of sterilization.

Unfortunately, in this study we were not able to examine in detail the method of sterilization. Although certain methods of sterilization have been shown to be more efficient in preventing subsequent pregnancy, the more efficient methods of preventing an intrauterine pregnancy are not always associated with a low frequency of tubal pregnancy. The more recent developments in the use of various clips may result in lower rates of intrauterine and ectopic pregnancies but, as more women at younger ages are sterilized with clips, there will be a longer time period during which an ectopic pregnancy might occur. Postpartum tubal ligation is now more likely to be undertaken in older parous women who, because of age-related declining fertility, have less chance of developing an ectopic pregnancy. This could bias assessments of the rates of ectopic pregnancy after sterilization.

The number of patients with ectopic pregnancy who have been sterilized can be expected to increase as we have confirmed, although the rate per 1000 sterilizations, considering all methods, remains relatively static. The increasing incidence of ectopic pregnancy, as expressed per 1000 sterilized woman-years, is a result of women being younger when sterilized. Passage of time is of limited protective value while ovulatory function persists and ectopic pregnancy must still be included in the differential diagnosis for women in the reproductive years presenting with acute abdominal pain irrespective of their sterilization status. Nevertheless, although important and increasing in number, examination of the data excluding previously sterilized women demonstrates that this contribution is small. The increasing incidence of ectopic pregnancy persists, and although infection must remain the prime suspect, other factors, such as

changes in contraceptive practice in the population, have an unknown effect.

The reason for the higher incidence of right-sided ectopic pregnancy is unknown. It may be due to secondary infection of the right tube after an appendicectomy or differences in referral patterns for investigation of right and left iliac fossa pain. It has recently been reported that ovulation tends to occur more frequently in the right ovary (Potashnik *et al.* 1987).

Salpingectomy was the treatment of choice in over 90% of patients. In the 1950s and 1960s removal of the ovary on the same side was more commonly practised to ensure that ovulation occurred on the side of the remaining tube, to facilitate ovum pick-up and to prevent transperitoneal migration of the ovum (Jeffcoate 1955). Conservative operations to remove an unruptured ectopic pregnancy, usually by linear salpingostomy or excision of the affected segment of tube, have risen from 0% in the 1950s to a current level of around 7%. The risks of post-operative haemorrhage and recurrence of an ectopic pregnancy in the conserved but damaged tube may have limited the popularity of such procedures.

In conclusion, we confirm an increasing incidence of ectopic pregnancy. A previous sterilization procedure does not exclude the diagnosis. It remains a serious and life-threatening condition and in the absence of careful practice, this rising incidence is likely to lead to an increase in maternal morbidity and mortality.

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Maternal blood viscosity and uteroplacental blood flow velocity waveforms in normal and complicated pregnancies

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Summary. According to the Poiseuille–Hagen law, viscosity influences flow resistance. A possible effect of blood viscosity upon the resistance index of the uteroplacental circulation as measured by continuous wave Doppler ultrasound was investigated in 50 pregnant women. It was found that blood viscosity variables explained only about 10% of the variation in the resistance index in all patients, which was not statistically significant. It is suggested, therefore, that the vascular contribution to flow resistance may be more important.

Pregnancies complicated by pre-eclampsia or intrauterine growth retardation or both are associated with Doppler flow velocity waveforms obtained from the uteroplacental circulation which demonstrate increased downstream resistance to flow (Campbell *et al.* 1983; Cohen-Overbeek *et al.* 1985). Such pregnancies are also associated with increased blood viscosity (Mathews & Mason 1974; Kaibara *et al.* 1981; Buchan 1982; Hobbs *et al.* 1982; Thorburn *et al.* 1982). The Poiseuille–Hagen law describes the relation between flow, pressure and resistance.

Expressed in a different form, resistance to flow is proportional to the fluid viscosity and inversely to the vessel radius to the fourth power:

$$\text{Resistance} \propto \frac{\text{Fluid viscosity}}{\text{Vessel radius}^4}$$

This study set out to determine the contribution of changes in blood viscosity to peripheral resistance in the uteroplacental circulation as determined by Doppler ultrasound.

Patients and methods

The study included 50 pregnant women of whom 15 demonstrated fetal growth retardation, diagnosed on ultrasound evidence of a fetal abdominal circumference of <5th centile on standard growth charts (Campbell 1974). Ten women had pre-eclampsia defined as a sustained blood pressure of $\geq 140/90$ mmHg with or without proteinuria in the second half of pregnancy but with a normal blood pressure at booking. Ten women had both pre-eclampsia and fetal growth retardation, and 15 had no evidence of either condition or any likely to be associated with impaired placental function. None of the women had diabetes. All were taking oral iron sup-

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plements but no other drugs at the time of testing.

Measurement of resistance in the uteroplacental circulation

The placenta was localized with a preliminary real-time ultrasound scan. The characteristic flow velocity waveforms were obtained from the placental side of the uterus with continuous wave Doppler ultrasound (Doptex, Chichester, UK) using a 4-MHz probe. After spectral analysis the waveforms were displayed on the screen and stored on audio tape. The mean resistance index (Pourcelot 1974) obtained from five ideal waveforms was used for the analysis. Waveforms were considered ideal if there was a high signal-to-noise ratio and the maximum frequency outline was clear.

Blood viscosity measurement

On the same day as the waveforms were recorded, 15 ml of maternal blood were taken from an antecubital vein without the use of a tourniquet, anticoagulated with potassium ethylene diamine tetra-acetic acid (K-EDTA), and blood viscosity and its determinants were measured within 4 h. Whole blood viscosity was measured at shear rates of 0.945 and 94.5 s⁻¹ using a Contraves LS30 rotational viscometer (Contraves Industrial Products Ltd, Ruislip, Middlesex) and plasma viscosity was measured using a Coulter-Harkness capillary viscometer (Coulter Electronics Ltd, Harpenden, Essex); both viscometers were operated at 37°C.

After centrifugation of the blood at 1500 g for 10 min, red cells were harvested from the centre of the packed cell column and suspended in phosphate-buffered saline (Dulbecco A, Oxoid Ltd, London) at approximately 25% haematocrit. The haematocrit of this suspension and of the whole blood was measured using a Coulter Counter (model S plus IV), and the red cell suspension was adjusted to 10%. Red cell filterability was assessed using a St George's Filtrimeter (Carri-Med Ltd, Dorking, Surrey) and 5-µm nuclepore filters from a single batch. The instrument measures the rate of flow of the red cell suspension through the filter at fixed intervals soon after the start of flow and automatically computes the initial rate of flow (Dormandy *et al.* 1985). From this measurement and the known haematocrit, the red cell transit time

(RCTT) is calculated; this variable characterizes the average resistance to flow of pores occupied by red cells compared to the resistance when suspending medium only is present. For the levels of residual white cells in the red cell suspension obtained in this study ($\leq 100/\mu\text{l}$), the RCTT value is independent of pore blockage by contaminating particles (Dormandy *et al.* 1985).

Statistical analysis

Means between the four groups were compared using Duncan's multiple range test. Rheological variables are known to vary throughout pregnancy (Buchan & MacDonald 1981; Buchan 1984), and the resistance index of the uteroplacental arteries decreases with increasing gestational age (Pearce 1987). It was therefore necessary to correct for gestational age in the analysis of the results. Relations between pairs of continuous variables were tested using the correlation coefficient, and the influence of gestational age on these relations was allowed for using multiple regression. The analyses were performed for the whole sample and for each individual subgroup.

Results

The gestational age, resistance index and viscosity measurements for each diagnostic group are displayed in Table 1. The mean resistance index in the group with both PE and IUGR was significantly higher than in the normal group or in the group with IUGR. The patients in the group with both PE and IUGR had a clinically more severe condition of earlier onset. In contrast, the patients with PE alone typically presented at a later gestation. The mean gestational age of these patients was significantly greater than that of the normal group and the group with both PE and IUGR. In accordance with previous reports, there is a consistent trend towards an increase in whole blood viscosity (at both high and low shear rates) across the patient groups, the lowest mean values were in the normal group, and the highest in the PE and IUGR group.

The main purpose of the statistical analysis was to determine whether blood viscosity or any of its determinants showed any consistent relation to the resistance index of the uteroplacental arteries. Table 2 shows the correlation coefficients between each blood viscosity variable and

Table 1. Summary statistics of each diagnostic group

	Normal (n = 15)	IUGR (n = 15)	PE (n = 10)	PE + IUGR (n = 10)	All complicated (n = 35)
Gestational age (weeks)	30.93 (4.27)	33.73 (4.04)	37.40* (1.58)	31.90 (4.82)	34.26 (4.27)
Resistance index	0.39 (0.07)	0.48 (0.17)	0.40 (0.12)	0.60** (0.18)	0.49 (0.17)
Whole blood viscosity at high shear rate (mPa.s)	4.03 (0.34)	4.13 (0.50)	4.17 (0.25)	4.38 (0.33)	4.21 (0.40)
Whole blood viscosity at low shear rate (mPa.s)	18.58 (4.39)	20.01 (4.75)	21.70 (3.12)	22.30 (4.94)	21.18 (4.38)
Haematocrit (%)	35.29 (2.28)	35.14 (4.10)	35.57 (2.21)	36.80 (2.72)	35.73 (3.27)
Plasma viscosity (mPa.s)	1.24 (0.05)	1.23 (0.07)	1.26 (0.06)	1.25 (0.09)	1.24 (0.07)
Red cell transit time	9.20 (0.82)	9.29 (0.59)	8.90 (0.55)	9.54 (1.02)	9.25 (0.75)

* $P < 0.01$ from the normal and (PE + IUGR) groups, ** $P < 0.01$ from the normal and IUGR groups.

Results are means (SD)

IUGR, Intrauterine growth retardation; PE, pre-eclampsia.

Table 2. Correlation coefficients (all patients)

	Resistance index	Gestational age
Whole blood viscosity at high shear rate	0.144	0.163
Whole blood viscosity at low shear rate	0.049	0.262
Haematocrit	0.105	0.142
Plasma viscosity	-0.060	0.187
Red cell transit time	0.297	-0.193
Resistance index		-0.342

the resistance index. This correlation was estimated for all viscosity variables in the 50 patients as an entire group, and also for each individual diagnostic sub-group. There was no significant correlation between resistance index and blood viscosity variables for any group. Figure 1 is a plot of resistance index against blood viscosity at low shear rate for all patients, which clearly demonstrates that the two variables bear no relation to one another. Figure 2 illustrates the range of viscosity results within each group and their relation to abnormal uteroplacental resistance results. A normal resistance index was considered to be less than 0.58 (Campbell *et al.* 1986).

From Table 2 it is clear that the viscosity variables and resistance index are related to gestational age. Since there was a significant difference between some groups in gestational age, statistical analysis allowed for this confounding variable. Table 3 shows the results of

multiple regression of resistance index and viscosity variables after gestational age has been taken into account. There was no significant relation between blood viscosity and resistance index in any group. For all subjects combined, blood viscosity variables could only explain a very small proportion of the variation in resistance index, which was not statistically significant.

Discussion

The Poiseuille-Hagen law describes the relation between flow, pressure and resistance:

$$\text{Flow} = \frac{\pi \Delta P r^4}{8L\eta}$$

where: r is the cylinder radius; ΔP is the pressure drop along the tube over a distance L , and η is the fluid viscosity.

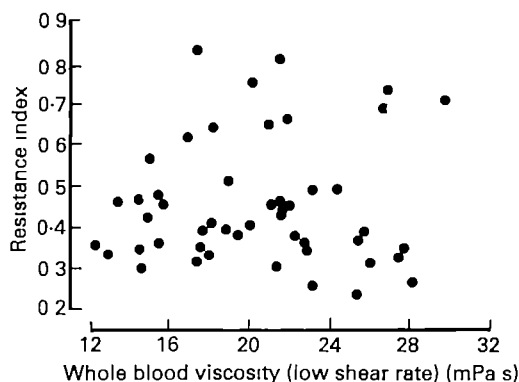


Fig. 1. A scattergram of resistance index against whole blood viscosity at low shear rate (0.945 s^{-1}) for all patients

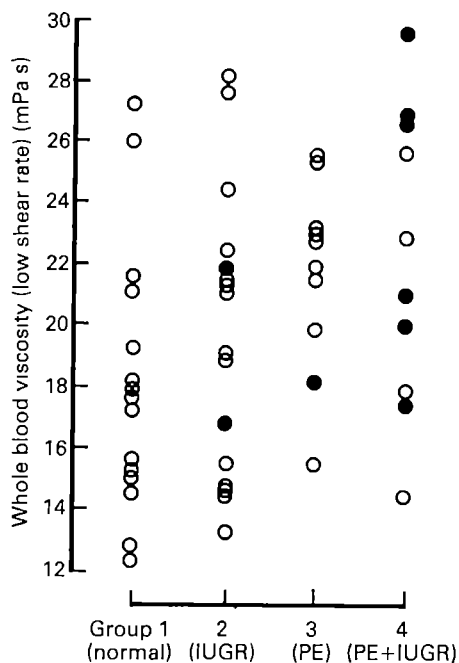


Fig. 2. The variation in whole blood viscosity at low shear rate (0.945 s^{-1}) within each pregnancy group demonstrating the distribution of normal (<0.58) and abnormal (≥0.58) resistance index values. IUGR, Intrauterine growth retardation; PE, pre-eclampsia. O, Resistance index <0.58; ●, resistance index ≥0.58.

This law applies to laminar flow of a Newtonian fluid in a straight rigid cylinder, and since blood is a non-Newtonian fluid this simple inverse relation between flow and measured blood viscosity may not necessarily apply to conditions *in vivo*. However, this relation has been verified by Dormandy (1971) with reference to blood flow in the human leg, and Greiss (1966)

demonstrated that it also applied to the circulation in a normotensive pregnant ewe.

The determinants of rheological resistance in the circulation are complex and vary according to vessel size. In capillaries, changes in red cell deformability have been shown to have a strong effect on blood flow velocity (Driessen *et al.* 1980). In vessels less than $150 \mu\text{m}$ in diameter, red cells migrate towards the centre of the vessel leaving a cell-free layer at the vessel wall. In these vessels plasma viscosity may be the major contributor to effective blood viscosity. In larger vessels the bulk viscosity as determined by rotational viscometry is a good measure of rheological resistance.

Our results of the mean values of whole blood viscosity at high and low shear rates and haematocrit in the different pregnancy groups agree with those of previous workers and demonstrate a trend towards increased viscosity values in the complicated pregnancies. We also found that the highest mean resistance index was recorded in the group with the most severe complications.

The greatest contribution of blood viscosity to the variation in resistance index was observed in patients with PE alone, where most patients demonstrated mild symptoms and signs. These patients, who all produced babies with weights appropriate for their gestational age, were therefore likely to have had an appropriate degree of plasma volume expansion (Campbell & MacGillivray 1977). This finding suggests that in these patients the rheological factor may have a greater influence than the vascular factor upon blood flow resistance compared with the other groups, but this would have to be confirmed by a study with a larger number of such patients.

In a similar study, Giles & Trudinger (1986) examined the possibility of an association between cord blood viscosity and Doppler measurement of umbilical artery resistance. They found a higher haematocrit in complicated pregnancies with high flow resistance than in those with low resistance; the difference was statistically significant. However, the effect of gestational age upon cord blood haematocrit (Buchan 1984) was not considered. They failed to find a significant association between abnormal umbilical artery flow resistance and cord blood viscosity at high or low shear rates.

The lack of correlation between blood viscosity and resistance index cannot be explained by inaccuracy in the viscometric measurements.

Table 3. Percent variability in resistance index explained by viscosity variables

	All patients (n = 50)	Normal (n = 15)	IUGR (n = 15)	PE (n = 10)	PE + IUGR (n = 10)
Whole blood viscosity (low shear rate)	2.0	0.0	4.0	15.0	3.0
Whole blood viscosity (high shear rate)	4.0	1.0	0.0	25.0	7.0
Haematocrit	2.0	7.0	0.0	36.0	20.0
Plasma viscosity	0.0	1.0	2.0	17.0	1.0
Red cell transit time	6.0	16.0	0.0	36.0	7.0
All viscosity variables	10.0				

In our hands, repeated measurements on individual samples showed a mean coefficient of variation of 2% (six samples each measured five times). Also, since blood was sampled on the same day that the resistance index was measured, only very small changes could occur in blood constituents between the measurements. A diurnal variation of about 10% has been found in blood viscosity (Ehrly 1986) although during the waking hours no significant variation has been detected (Ernst *et al.* 1985). Continuous measurement of resistance index each minute over a 30-min period in seven women at rest showed a mean coefficient of variation of 9%.

The absence of a relation between uteroplacental flow resistance and blood viscosity (and its determinants) in the present study suggests that changes in vascular architecture are more important. During normal pregnancy, extravillous trophoblasts invade the spiral arteries, converting them to uteroplacental arteries which are devoid of a muscular coat and probably unresponsive to pressor agents. In pre-eclampsia and to a lesser extent in fetal growth retardation this physiological response is impaired (Sheppard & Bonnar 1976; Brosens *et al.* 1977). The approximate diameter of a uteroplacental artery with normal trophoblastic invasion is 500 µm, compared to 200 µm when there is no such response (Brosens *et al.* 1972). If all other factors are equal, trophoblastic invasion leads to a 39-fold decrease in the resistance to flow in uteroplacental arteries according to the Poiseuille-Hagen law. This change in vascular resistance may be expected to outweigh the contribution to flow resistance of relatively small changes in blood rheology. Examination of placental bed biopsies taken at the time of caesarean section in women with severe pre-eclampsia shows that the absence of tropho-

blastic invasion correlates well with an abnormally high resistance index (Pearce 1987).

In summary, although certain abnormalities of blood rheology are more likely in patients with fetal growth retardation, pre-eclampsia, or particularly both conditions, small changes in viscosity do not appear to influence downstream resistance to flow in the uteroplacental arteries as determined by continuous wave Doppler ultrasound studies. It seems more likely that the impairment of uteroplacental blood flow in these conditions is associated with vascular rather than rheological abnormalities.

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Serial fetal blood sampling for the management of pregnancies complicated by severe rhesus (D) isoimmunization

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Summary. Fifty-one pregnancies complicated by rhesus (D) isoimmunization have been managed by serial fetal blood sampling between 17 and 36 weeks gestation as an alternative to amniocentesis for ΔOD^{453} measurements. In 36 pregnancies where the fetus was shown to be rhesus (D) positive and both measurements were made before any intrauterine fetal transfusions, the ΔOD^{453} value gave misleading predictions on 13 of 63 occasions (21%). Fetal haematocrit estimations provided a direct assessment of the haemopoietic compensation occurring, but fetal bilirubin and albumin concentrations did not correlate directly with disease severity. It is proposed that pregnancies complicated by severe isoimmunization can be more precisely managed by serial fetal blood sampling for haematocrit estimation than amniocentesis for ΔOD^{453} measurement thus avoiding unnecessary intervention or delayed treatment.

Despite the widespread use of immunoglobulin prophylaxis, pregnancy losses still occur from rhesus (D) isoimmunization (Clarke *et al.* 1985; Bowell *et al.* 1985). The most widely used method to assess the severity of isoimmunization is serial measurements of amniotic fluid ΔOD^{453} , proposed by Liley (1961). The timing of amniocentesis has usually been made by observing trends in maternal serum anti-D levels with con-

sideration of the maternal history of previous rhesus sensitized pregnancies. Several centres have found that ΔOD^{453} measurements give an incorrect prediction of fetal condition in 5–20% (Pridmore *et al.* 1972; Fairweather *et al.* 1976; Bowman 1978; Weiner *et al.* 1981).

MacKenzie *et al.* (1983) proposed that fetal blood sampling (FBS) should be used to determine the fetal rhesus type in those pregnancies where the father is heterozygous for rhesus (D) and there is a significant threat from high circulating antibodies. We have now studied pregnancies where the fetus is known or expected to be rhesus (D) positive, to see whether serial FBS will give a more accurate assessment of fetal haemolysis/anaemia than amniocentesis and will readily identify those fetuses in whom severe anaemia or hydrops can be expected.

Patients and methods

Fifty-one pregnancies associated with high or rising anti-D antibodies in the maternal circula-

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tion were investigated by FBS. The first procedure was performed after 17 weeks gestation if the anti-D potency was ≥ 10 i.u./ml except in those patients with a history of a previous pregnancy affected by severe rhesus disease, who may have been sampled if the potency was ≥ 4 i.u./ml. Most FBS procedures used fetoscopy (MacKenzie & Maclean 1980) and more recently an ultrasound-guided needle technique (M. Selinger *et al.*, unpublished observations).

Blood was drawn through dry needles into dry syringes for haematocrit estimation, fetal blood group and Kleihauer analyses and into heparinized syringes for bilirubin and albumin measurements. At the same time, an aliquot of amniotic fluid was collected into a darkened

glass tube for ΔOD^{453} estimation using Liley's method of interpretation; all samples with any overt blood contamination were routinely centrifuged immediately after collection. The subsequent management of the pregnancy was determined by the fetal blood results with regard for the continuing maternal anti-D potency and period of gestation. Further FBS was performed usually at 2–4-week intervals, the more frequent interval if the previous fetal haematocrit result was near our lower limit for normal, or showed a downward trend, or there was a significant ($>50\%$) increase in maternal anti-D potency after the FBS; no samplings were performed after 36 weeks gestation. Intravascular fetal transfusions were given if the fetal haematocrit was $\leq 25\%$ up to 25 weeks gestation and $\leq 30\%$ at 25–35 weeks gestation; data collected subsequently from these patients were not included in the analyses. The decision to deliver the woman was usually made between 32 and 36 weeks gestation depending on fetal condition.

To obtain normal values for fetal blood during pregnancy, samples were collected at fetoscopy from non-rhesus immunized pregnancies and analysed for haematocrit, bilirubin, and albumin.

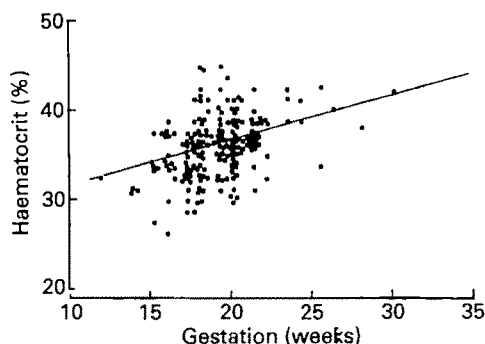


Fig. 1. Fetal haematocrit values according to gestation in 160 non-sensitized pregnancies showing a regression line.

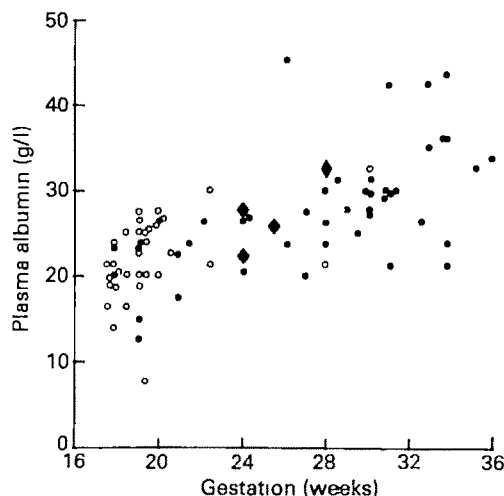


Fig. 2. Plasma albumin concentrations in fetal blood according to gestation. ○, Non-sensitized pregnancies; ●, rhesus isoimmunized pregnancies; ◆, four hydropic rhesus immunized fetuses.

Results

All blood samples were confirmed by the Kleihauer test to be of fetal origin, uncontaminated by maternal blood.

The fetal haematocrit values for non-sensitized fetuses are shown in Fig. 1; although there was a wide variation over the gestational period studied, there was a significant correlation between gestation and haematocrit ($r=0.28$; $T=3.59$; $n=153$; $P<0.0001$). Figure 2 shows the fetal plasma albumin levels in the non-sensitized pregnancies and in the rhesus isoimmunized pregnancies. There was a significant correlation for the non-sensitized pregnancies between albumin concentration and gestation ($r=0.38$; $T=2.38$; $n=33$; $P<0.02$); a similar trend was observed for the rhesus pregnancies ($r=0.47$; $T=3.63$; $n=49$; $P=0.0007$).

Bilirubin concentrations were not influenced by gestation in the sensitized and non-sensitized pregnancies. The maximum value obtained in 35 non-sensitized pregnancies was $50 \mu\text{mol/l}$, mean 23.7 (SD 12.0) $\mu\text{mol/l}$, compared with a maximum of $135 \mu\text{mol/l}$ and mean 68.5 (SD 32.8) $\mu\text{mol/l}$ for the sensitized pregnancies ($P<0.001$,

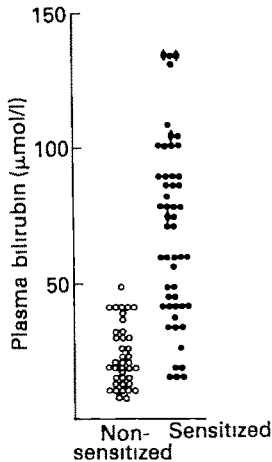


Fig. 3. Fetal plasma bilirubin concentrations in non-immunized (○) and rhesus immunized pregnancies (●). Four hydropic fetuses are identified as ◆

Student's *t*-test) (Fig. 3). There was no correlation between fetal haematocrit and bilirubin concentration in either the sensitized or non-sensitized pregnancies.

Of the 51 pregnancies complicated by rhesus (D) isoimmunization, 11 fetuses were found to be rhesus (D) negative and these were not sampled again. One of these 11 pregnancies miscarried 1 month after sampling at 18 weeks gestation and the other 10 babies were born healthy and survived; two were born preterm.

Of the 40 pregnancies where the fetus was rhesus (D) positive, one was terminated and one was managed elsewhere; the baby was born alive and healthy at 35 weeks gestation after amniocentesis at 34 weeks. Twenty of the remaining 38 pregnancies were managed by FBS alone on one to four occasions over periods of up to 12 weeks until delivery; the other 18 required intrauterine intravascular transfusions, 10 at the time of the initial sampling or within 3 days, and eight following a second, third or fourth sampling procedure.

Maternal anti-D quantitation was greater than 20 i.u./ml in 16 of the 18 pregnancies where the fetus was anaemic (haematocrit <25% by 25 weeks gestation or <30% after that gestation) at the first or subsequent blood sampling. In the other two, repeated maternal plasmapheresis had been performed, which halved the original anti-D concentration to 16 and 15 i.u./ml respectively.

In 36 pregnancies both ΔOD^{453} and fetal haematocrit were measured on a total of 63 occa-

sions. There was good agreement between the two tests in 50 (79%) instances. There were 13 occasions during 12 pregnancies, however, when there were discordant results (see Fig. 4). In seven (11%) the ΔOD^{453} value was in Liley's zone II (or the backward extrapolation before 28 weeks gestation) and gave false reassurance about the fetal condition when the fetal haematocrit was low and transfusion or delivery was indicated; in four this was before and in three after the 28th week. In six further instances (10%), all in the third trimester, the ΔOD^{453} value was falsely alarming in Liley's zone III when the fetal haematocrit was above 30% in four and above 40% in two; all these pregnancies were managed by further sampling rather than fetal transfusion or delivery.

In seven of the 40 (17%) rhesus (D)-positive pregnancies, the haematocrit was between 30% and 40% at the first sampling at 18–30 weeks gestation and all were managed by serial FBS rather than immediate fetal transfusion. The number of samplings per pregnancy ranged between two and four and the interval between

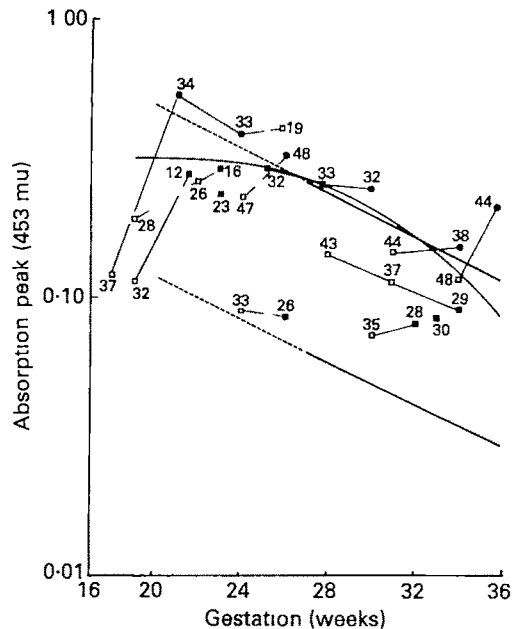


Fig. 4. Discordant fetal haematocrit and amniotic fluid ΔOD^{453} values in 12 pregnancies managed by repeated fetal blood samplings. Individual points show matched haematocrit and ΔOD^{453} values with ● denoting false-positive and ■ false-negative ΔOD^{453} results. Also illustrated are Liley zone lines (Liley 1961) and the 'Whitfield action line' (Whitfield 1970).

the first sampling and delivery or fetal transfusion was between 3 and 7 weeks.

In four pregnancies, the fetus was hydropic at the time of referral to the unit at 24–28 weeks gestation. The fetal haematocrit values were 9%, 12%, 25% and 29% respectively; the plasma albumin and bilirubin levels are identified in Figs 2 and 3. There was no indication that these four hydropic fetuses had abnormally low plasma albumin levels. None of these four fetuses survived. Six other fetuses that were transfused *in utero* did not survive to delivery, and eight were born alive and subsequently discharged from hospital.

Of the 20 pregnancies monitored by serial FBS without intrauterine transfusion, 18 resulted in healthy infants, three were born after spontaneous preterm labour at 27–35 weeks, one of whom did not survive the neonatal period. There was one intrauterine fetal death, three days after sampling associated with placental abruption and one baby was born electively very preterm for severe intrauterine growth retardation and died a few hours after birth.

Discussion

Most units managing pregnancies complicated by rhesus haemolytic disease rely upon amniocentesis and ΔOD^{453} measurements, a surrogate test assessing fetal haemolysis. However, there have been several reports of false predictions both positive and negative in the fetal condition in up to 20% of pregnancies where this approach was used. The present results endorse this view.

Alternative methods of management have been explored including cardiotocography to detect the development of sinusoidal cardiac rhythms (Rochard *et al.* 1976; Visser 1982) and measurement of intraventricular conduction time (Brambati & Pardi 1981). Fetal movements have been assessed clinically (Sadovsky *et al.* 1979) and by ultrasound (Frigoletto *et al.* 1986) while other sonar observations including the detection of hydropic changes (Weiner *et al.* 1981) and the measurement of umbilical vein diameter (DeVore *et al.* 1981) and blood flow (Kirkinen *et al.* 1983) have also been advocated. Where an objective assessment has been made, each has less than 100% specificity and sensitivity and almost all will detect severe disease only when fetal and neonatal survival rates are already reduced.

Fetal blood sampling, as proposed, provides a

direct assessment of fetal compromise by determining the degree of haemopoietic compensation to haemolysis. The fetal response to red cell sensitization and resultant haemolysis is variable and although others have claimed to the contrary, we believe that the development of hydrops fetalis does not correlate precisely with fetal haematocrit (Nicolaidis *et al.* 1986). However, we have not seen hydrops when the haematocrit was above 30% and only once above 25%. Similarly we have found no evidence of hypoalbuminaemia in fetuses suffering from hydrops fetalis as reported by others in neonatal observations (Phibbs *et al.* 1974; Bowman 1978; DeVore *et al.* 1982). We acknowledge, nevertheless, that the samples from non-sensitized fetuses were limited in the third trimester for comparison: we can see no way at present of obtaining many samples from 'normal' pregnancies at a late gestation to provide a library of 'normal' values.

We are unable to prove conclusively that an earlier intervention (transfusion or delivery) was justified on the fetal blood results when the ΔOD^{453} value was in Liley's zone II, although in two pregnancies the haematocrits were 12% and 16% respectively and the fetuses thus severely anaemic. We have convincing evidence, however, that 11% of patients avoided unnecessary intrauterine transfusions or very early delivery when the ΔOD^{453} values were in Liley's zone III and the fetal haematocrit was above 33%. We further believe that fetal transfusions were avoided or delayed in 6 (17%) by serial monitoring of fetal haematocrit levels when the value was between 30 and 40%. We suggest that the approach advocated by some workers of transfusing the fetus when the haematocrit is less than 40% is unnecessarily aggressive (Rodeck *et al.* 1984).

Although we have not fully assessed the morbidity associated with FBS compared with amniocentesis due to the relatively small numbers studied, we believe that the two techniques probably carry similar risks, a view expressed by others (Rodeck & Nicolaidis 1983). We are nevertheless concerned about the increase in maternal antibody concentration that occurs after sampling especially when the placenta is traversed using ultrasound-guided needle sampling; this aspect of blood sampling is currently under investigation (Bowell *et al.* 1988).

Using a direct assessment of the fetal con-

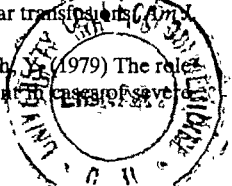
dition measuring fetal haematocrit, unnecessary intrauterine transfusions and preterm deliveries have been avoided and we have reduced the frequency of intrauterine investigations. We have found no reason to repeat sampling within 1 week and in general we repeat sampling at 2–4 week intervals, thus comparing favourably with amniocentesis which some advocate repeating once or twice weekly (Bowman 1978). However, FBS is invasive and we firmly believe that strict criteria should be observed before subjecting a pregnancy to sampling. Irrespective of previous history, we consider it inappropriate to perform FBS unless maternal anti-D concentrations are above 10 i.u./ml between 18 and 36 weeks gestation and not as low as 4 i.u./ml as recommended by Nicolaides *et al.* (1986). Indeed, we observed no anaemic fetuses (haematocrit <30%) when the maternal antibody potency was less than 20 i.u./ml (except when repeated maternal plasmapheresis was being performed); our threshold for intervention may thus have to be modified further. In order to obtain maximum fetal salvage, we recommend that severely compromised rhesus (D) pregnancies should be referred to a unit where blood fetal sampling can be performed.

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Antenatal fetal blood sampling for the management of alloimmunized pregnancies: effect upon maternal anti-D potency levels

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Summary. Increase in maternal anti-D concentrations after intrauterine investigation has been studied retrospectively in 95 rhesus (D) alloimmunized pregnancies; 48 were managed by fetal blood sampling (FBS) procedures (using fetoscopy or ultrasound-guided needle sampling) and 47 using amniocentesis. In those pregnancies where the fetus was rhesus (D) positive, the frequency of procedure-related increases ($>50\%$) in maternal anti-D potency was estimated following single procedures and found to be similar for the two methods of FBS employed (28%) and for amniocentesis (31%). The proportion of pregnancies showing an increase in anti-D potency was higher after ultrasound-guided needle sampling (75%) than after fetoscopic FBS (40%) and after amniocentesis (44%).

Fetal blood sampling (FBS) is gaining acceptance in the management of severe haemolytic disease of the newborn (HDN), to determine fetal rhesus(D) type (MacKenzie *et al.* 1983), to assess fetal response to haemolysis (Nicolaidis *et al.* 1986; MacKenzie *et al.* 1988), and an extension of the procedure is used for direct intravascular transfusion (MacKenzie *et al.* 1982; Rodeck *et al.* 1984; Selinger *et al.* 1987).

Although amniocentesis is known to be capable of provoking feto-maternal haemorrhage

(FMH) (Medical Research Council 1978; Dubin & Staisch 1982), little is known about the prevalence of FMH after fetal blood sampling. To assess this possible hazard, we investigated the associated changes in maternal anti-D potency after FBS procedures and after amniocentesis.

Patients and methods

Between 1982 and 1986, 95 pregnancies at risk of moderate to severe rhesus(D) HDN, as judged either by maternal anti-D potency values or from a history of previous severe HDN, were seen in the Oxford Region. Forty-eight pregnancies were referred to the John Radcliffe Hospital, Oxford, for investigation and treatment using FBS procedures; 47 pregnancies managed by amniocentesis, performed by local obstetricians in 10 district general hospitals, served as non-randomized controls.

The type of management each patient received depended upon maternal anti-D potency, gestational age and the distance that the patient lived from Oxford. Women with

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higher anti-D levels in early pregnancy tended to be investigated more often by FBS procedures because of the greater chance of fetal transfusion being needed.

FBS was initially performed under direct vision by fetoscopy as described previously (MacKenzie & MacLean 1980) and more recently using an ultrasound-guided needle technique (Daffos *et al.* 1983; Selinger *et al.* 1987); the first sampling was generally not performed until the maternal anti-D potency had reached 10 i.u./ml. Both procedures were used in some pregnancies but not on the same occasion. The fetoscopic method was generally preferred for sampling alone and the ultrasound-guided method for intravascular fetal transfusion (MacKenzie *et al.* 1987; Selinger *et al.* 1987). Maternal blood samples were examined before and after the procedure for evidence of FMH using Shepard's modification (Shepard *et al.* 1962) of the Kleihauer technique (Kleihauer *et al.* 1957): results were reported as the number of fetal cells seen per 50 low-power microscope fields.

Amniocentesis was performed under the direction of the local obstetrician with consideration of our previous advice that whenever possible the procedure should not be used if the maternal anti-D potency remained below 4 i.u./ml (Bowell *et al.* 1982). Details of the procedures used, including the use of ultrasound guidance to avoid placental trauma, were not provided, and neither were Kleihauer results available.

Automated anti-D quantitation procedures were carried out as described previously by Bowell *et al.* (1982) using the British National anti-D standard preparation coded 72/229 for comparative assay. The maternal anti-D concentrations in blood samples taken immediately before an intrauterine procedure were compared with those obtained 2 weeks before and 2 weeks after that procedure. Occasional samples with the automated method show an inherent variability in the region of $\pm 25\%$ (Gunson *et al.* 1974), but for most samples the overall level of reproducibility lies within $\pm 10\%$; thus a rise in maternal anti-D of more than 50%, twice the level of poorest reproducibility, was judged to be significant (Bowell *et al.* 1982). To avoid including the results of spontaneous sensitizations, data from pregnancies in which there were significant rises during the 2 weeks preceding the procedure were excluded from the analysis; data

from pregnancies which did not continue for a further 2 weeks after the procedure were also excluded.

Estimates of the frequency of procedure-related increases ($>50\%$) in maternal anti-D levels, for those pregnancies where the fetus was rhesus(D) positive, were made both after individual procedures during a pregnancy and overall for each pregnancy.

Results

A total of 96 FBS procedures was performed during the 48 pregnancies in the FBS group. The number of procedures per pregnancy ranged from 1 to 6 and these were carried out between 17 and 35 weeks gestation. The first FBS was performed on average at 24 weeks gestation when the mean anti-D potency was 56 i.u./ml (range 4.0–725 i.u./ml). Fetoscopy was used on 68 occasions, 53 (78%) for blood sample collection alone and 15 for intravascular fetal transfusion. The ultrasound-guided procedure was used on 28 occasions, 10 (36%) for sampling and 18 for fetal transfusion.

Amniocentesis was performed on 90 occasions in the other group of 47 pregnancies with a range of 1–5 procedures per pregnancy. The first amniocentesis was carried out on average at 32 weeks gestation (range 26–37 weeks) when the mean anti-D potency was 15 i.u./ml (range 0.51–89 i.u./ml).

In the group of pregnancies managed with FBS procedures, 11 fetuses were found to be rhesus(D) negative at the initial procedure. All pregnancies continued for at least 2 weeks and no maternal serum showed a rising anti-D potency. In the amniocentesis group there were five rhesus(D)-negative babies and no maternal serum showed a rising anti-D potency.

Frequency of increased anti-D potency after one procedure

Details of procedure-related increases in anti-D levels for those pregnancies where the fetus was rhesus(D) positive are shown in Table 1. Of the 47 FBS and 36 amniocenteses studied, the proportion associated with a rise of anti-D after the procedure was similar, 28–31%, for all three investigation methods. Of the eight increases associated with fetoscopy, one occurred after fetal transfusion and seven after sampling alone; with the ultrasound-guided method one rise

occurred after fetal transfusion and four after sampling.

Frequency of pregnancies showing procedure-related increases

Of the 37 pregnancies managed by FBS where the fetus was rhesus(D) positive, the anti-D potency of 14 could not be monitored for 2 further weeks after one or more procedures. Increases after the procedure (>50%) in anti-D potency were seen in 13 of the 23 (57%) remaining pregnancies (Table 2). Of 25 pregnancies managed by amniocentesis 11 (44%) showed significant increases. Pregnancies where ultrasound-guided procedures were used showed a greater frequency (75%) of post-procedure rises than those managed by fetoscopy (40%); all four

pregnancies (100%) sampled by both procedures showed significant increases. Although the average number of procedures per pregnancy was greatest for those where the ultrasound-guided method was used, either alone or in addition to fetoscopy, on five of the seven occasions where an increase in potency was observed it was found to occur after the initial sampling. None of the pregnancies sampled more than once showed more than one major rise (>50%) in maternal anti-D potency.

An assessment of the average anti-D potency increments during a pregnancy was made for each sampling method by contrasting the highest anti-D level after the procedure with the potency level before the initial sampling (H:L ratio). The mean values for each sampling method show (Table 2) that higher ratios were seen in those

Table 1. Changes in maternal anti-D potency within 2 weeks of a procedure (in pregnancies where the fetus was rhesus(D) positive), showing frequency of a significant increase (>50%) after the procedure

Procedure	Excluded from the analysis		Included in the analysis	
	Increase before procedure	Unable to monitor potency 2 weeks later	Potency monitored 2 weeks later	Significant increase after procedure
Fetoscopy (<i>n</i> =57)	8	20*	29	8 (28%)
Ultrasound-guided needle sampling (<i>n</i> =28)	5	5†	18	5 (28%)
Amniocentesis (<i>n</i> =82)	14	32‡	36	11 (31%)

* Intrauterine death = 8; preterm delivery = 7; termination = 1; transferred = 2; further fetal transfusion within 2 weeks = 1; miscarriage = 1.

† Intrauterine death = 2; preterm delivery = 1; further fetal transfusion within 2 weeks = 2.

‡ Preterm delivery = 25; further amniocentesis within 2 weeks = 7

Table 2. Frequency of pregnancies (where fetus was rhesus(D) positive) showing procedure-related increases (>50%) in maternal anti-D levels, indicating comparative mean anti-D potency values for each investigation technique in pregnancies that showed a significant increase

Procedure	Anti-D* potency monitored	Significant (>50%) rise in potency	Procedures/pregnancy	Mean values for pregnancies showing >50% rise		
				Anti-D level (i.u./ml)		High:low (H:L) ratio
				At first sampling	Maximum	
Fetoscopy (<i>n</i> =28)	15	6 (40%)	2.5	28	155	5.5:1
USGN (<i>n</i> =5)	4	3 (75%)	4.0	14	252	18:1
Fetoscopy+USGN (<i>n</i> =4)	4	4 (100%)	5.0	28	579	21:1
Amniocentesis (<i>n</i> =42)	25	11 (44%)	2.6	11	108	10:1

* Remainder not included because one or more of the following occurred within 2 weeks of a procedure. preterm delivery, intrauterine death, rise in anti-D levels before the procedure, transferred elsewhere for management, further procedure within 2 weeks, miscarriage, termination of pregnancy.

USGN, Ultrasound-guided needle sampling.

pregnancies managed by the ultrasound-guided method alone (H:L ratio 18:1) or in addition to fetoscopy (H:L ratio 21:1) than after fetoscopy alone (H:L ratio 5.5:1) and after amniocentesis (H:L ratio 10:1).

No fetal cells were seen in the Kleihauer tests in any of the 63 maternal bloods examined before the procedure and in 66 of the 72 samples tested after the procedure. A positive Kleihauer result was seen after four of the 12 ultrasound-guided samplings and two of 60 fetoscopic procedures ($P < 0.005$, χ^2) (Table 3). Of these six pregnancies which produced rhesus(D)-positive infants, four continued for 2 weeks after the relevant procedure, but only one maternal serum showed a rising anti-D level (Kleihauer = 74 fetal cells per 50 low-power microscope fields). Although two of these four had had a significantly rising anti-D level after a previous procedure, neither showed an additional increment at this time or during the remaining 9 weeks of pregnancy when there were three further procedures. Kleihauer tests were performed after eight of the 13 procedures where there were procedure-related increases in potency; seven showed no fetal red cells in the maternal circulation.

Discussion

Recently developed obstetric procedures combined with intensive paediatric management have improved the prognosis in pregnancies severely affected by rhesus disease especially during earlier gestation. Intrauterine FBS procedures allow a more precise assessment of fetal condition and can be used for direct intravascular transfusion (MacKenzie *et al.* 1982; Rodeck & Nicolaides 1983; Nicolaides *et al.* 1986; Mac-

Kenzie *et al.* 1987). Although these procedures are known to carry the risk of miscarriage, amniotic fluid leakage and premature labour (Rodeck & Nicolaides 1983), we have found no previous objective assessment of the influence of FBS on maternal antibody stimulation.

For this study, the automated anti-D quantitation method was preferred to the manual titration technique because of its improved precision and reproducibility. It is known that with this method 68% of maternal sera exhibit an overall rise in anti-D potency of more than 50% during pregnancy when the fetus is rhesus(D) positive (Bowell *et al.* 1982), but it is not clear whether this increase relates to obstetric procedures or occurs spontaneously. Although we endeavoured not to overestimate the frequency of procedure-related antibody stimulation by excluding spontaneous sensitizations, it was possible using our protocol that this frequency was underestimated because: not all secondary sensitizations are revealed within 2 weeks (Mollison 1983), data from pregnancies which did not continue for 2 further weeks were excluded from the study, and small increases in anti-D potency (<50%) were disregarded.

The frequency of significant increases in anti-D potency after the procedure appeared to be similar for the three investigation methods (Table 1). It was apparent, however, that the hazards of the ultrasound-guided procedure were underestimated because when multiple sampling was required, a significant increment was most commonly seen (five of seven occasions) after the initial procedure and no further increases occurred after subsequent samplings in those pregnancies. Although anti-D assays were not performed during the postnatal period, it is assumed that these patients had probably

Table 3. Kleihauer test results on maternal blood samples following fetal blood sampling

Procedure	Application	Kleihauer test results after the procedure (cells per 50 low-power microscope fields)				
		NT	0	1-10	10-50	>51
Fetoscopy	Sampling ($n=53$)	7	44	1	1	0
Fetoscopy	IVT ($n=15$)	1	14	0	0	0
USGN	Sampling ($n=10$)	4	3	1	0	2
USGN	IVT ($n=18$)	12	5	0	0	1

NT, Not tested.

USGN, Ultrasound-guided needle sampling.

IVT, Intravascular fetal transfusion.

reached maximum antibody production levels after the first sampling procedure and thereafter could not further increase anti-D output.

The 31% frequency (Table 1) of rising antibody levels observed after amniocentesis was higher than the 10% average estimate of FMH reported by others using the Kleihauer method (Dubin & Staisch 1982). It is known, however, that not all FMH are shown by the Kleihauer technique and 5% of patients experience a primary immune response to rhesus(D) at delivery when no cells are seen in the maternal circulation immediately postpartum (Woodrow *et al.* 1965). It is likely that secondary sensitization, which is thought to require a smaller antigenic challenge (Mollison 1983), would occur more frequently than primary sensitization. The frequency of increasing anti-D levels seen after amniocentesis might also be higher than expected because we have used pooled data from a number of hospitals, and it has previously been shown that whereas individual obstetricians are able to attain high levels of expertise in amniocentesis, the overall abilities of many practitioners will exhibit wide variations (Medical Research Council 1978; Dubin & Staisch 1982).

The relatively low prevalence of fetoscopy-related anti-D increases is shown by both the comparative chances of antibody stimulation at a single procedure (Table 1) and the overall rate per pregnancy (Table 2). The average anti-D increases associated with fetoscopy are also the lowest of the three sampling methods. It is possible that this relatively low risk might be attributable to the more frequent use of ultrasound guidance for fetal transfusion, which is a longer intrauterine procedure. However, four of five rises related to the ultrasound-guided procedure occurred after sampling alone, as did seven of eight increases associated with fetoscopy. A more likely explanation for the difference lies within the techniques themselves: in fetoscopy procedures there is an effort to avoid placental trauma but with the ultrasound-guided method there is usually passage of the needle through the body of the placenta into the umbilical cord insertion (Daffos *et al.* 1983; Selinger *et al.* 1987).

Kleihauer tests after the procedure possibly provide indications of a large FMH (Queenan 1977), but appear to give little or no indication of the smaller FMH which can provoke antibody stimulation. Of the eight occasions associated

with an anti-D rise after the procedure, only one maternal sample showed any fetal cells in the Kleihauer test. Possibly, in the presence of high maternal anti-D levels, most fetal cells are rapidly removed from the maternal circulation and are not seen on Kleihauer slides, but are capable of stimulating anti-D production. The presence of fetal red cells in the maternal circulation, however, does not always indicate an impending increase in anti-D, since only one of four women subsequently had a rising anti-D level after a positive Kleihauer test after the procedure.

It is probable that a significant rise in maternal anti-D levels accelerates fetal haemolysis and results in earlier planned delivery or more extensive neonatal treatment than might otherwise have been necessary. In the more severe forms of rhesus(D) HDN, where intrauterine transfusion is necessary, it could be argued that following transfusion of the fetus with rhesus(D)-negative blood, the maternal anti-D potency is of limited importance. We submit, however, that a raised maternal anti-D level may lead to a requirement for more intensive fetal transfusion therapy, very high fetal plasma anti-D levels which can require prolonged neonatal therapy, and in some instances high starting anti-D levels in subsequent pregnancies thereby possibly worsening the prognosis.

This report of a non-randomized retrospective study, correlating rises in maternal anti-D levels with two assessment methods, emphasizes the requirement for careful selection of patients for investigation and treatment. We have long held this view regarding the implementation of amniocentesis (Bowell *et al.* 1982). With FBS procedures, however, the problems are more acute given both the earlier application of the technique and the relatively high levels of anti-D attained.

We recommend that FBS should only be used when either the maternal anti-D potency reaches significant levels according to local criteria or when there is a history of previous severe HDN; our current minimum maternal anti-D level for intervention has been set at 10 i.u./ml. In our experience the anti-D potency reaches this level in only 15% of sensitized patients, and two-thirds of these do so after 28 weeks gestation when early delivery might be considered.

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Studies on the effects of nitrendipine on oxytocin-, angiotensin II- and ergometrine-induced contraction of pregnant human myometrium *in vitro*

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Summary. Calcium channel blockers inhibit myometrial contractility by preventing the increase in intracellular free calcium which follows stimulation. They could thus be useful in treating premature labour. The effect of nitrendipine, a dihydropyridine calcium channel antagonist, on the contractile response of strips of pregnant human myometrium to oxytocin, angiotensin II (AII) and ergometrine has been examined. A total of 68 tissue strips were studied, with random allocation to treatment group. Initial concentration: response curves to one of the three agonists were determined; the concentration: response determinations were repeated in the presence or absence of nitrendipine at 10^{-9}M . The initial EC_{50}s for tissues exposed to oxytocin and AII were $8.2 \times 10^{-10}\text{M}$ and $3.4 \times 10^{-8}\text{M}$ respectively. The contractile response to both agonists was significantly blunted in the presence of nitrendipine (ANOVA; $P < 0.001$ for both agents). This effect was greatest at high agonist concentrations. The initial EC_{50}s for tissues exposed to ergometrine was $3.9 \times 10^{-8}\text{M}$. Exposure to nitrendipine blunted the response (ANOVA; $P < 0.001$), an effect most marked at low concentrations of ergometrine. The effect of nitrendipine on myometrial responses to the naturally occurring hormones oxytocin and AII supports suggestions of a role for it in inhibiting premature labour.

Myometrial contraction, like that of vascular smooth muscle, is initiated by an increase in the concentration of intracellular free calcium (Bolton 1979) which forms a complex with calmodulin. This activates a kinase which in turn catalyses the phosphorylation of myosin, allowing its interaction with actin. Myometrial

smooth muscle appears primarily to be activated by the influx of calcium from the extracellular fluid, rather than by the release of intracellular stores (e.g. Maigaard *et al.* 1986a). Thus, agents which block the entry of calcium into the cells might be expected to inhibit myometrial contraction and hence suppress premature labour.

In the past few years, some calcium antagonists have been found to be effective inhibitors of stimulated myometrial contraction in non-human species (e.g. Grosset & Mironneau 1977) and in man (e.g. Ulmsten *et al.* 1978, 1980; Maigaard *et al.* 1986a). Their use in the treatment of premature labour seems logical, and might avoid the need to use powerful sympathomimetic agents. We report the effect of nitrendipine, a

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dihydropyridine calcium antagonist, on induced contractions of pregnant human myometrium *in vitro*, using oxytocin, angiotensin II (AII) and ergometrine maleate as agonists. The choice of agonists has been influenced by the therapeutic role of oxytocin and ergometrine in the management of labour, and the possible role of AII in the physiology and pathophysiology of pregnancy.

Materials and methods

The myometrial strips were obtained from women undergoing caesarean section. The tissues were excised in the line of the edges of the lower segment incision and were immediately placed in Krebs' solution containing $11.1 \times 10^{-3} \text{ M}$ glucose (Kitson & Broughton Pipkin 1981) and transferred to the laboratory. Strip preparations approximately 2–5 mm wide, 0.5–1.0 mm thick and 20–38 mm long were dissected out. At least four myometrial tissue strips were prepared from each uterus. The strips were mounted in modified individual 5-ml tissue baths (Odum & Broughton Pipkin 1987). During wash-out periods the Krebs' solution in the tissue baths was continuously changed at a rate of 0.1 ml/min and maintained at 37 °C throughout. Contractions were recorded isotonicly against a tension of 500 mg, using Ferranti 8 HL1 micro-torque potentiometers (Ferranti plc, Scotland).

The tissues were left to equilibrate for at least 60–90 min after setting up. The response to 100 mM KCl (final concentration) was then determined to ascertain tissue viability and the tissues were subsequently allowed to re-equilibrate in Krebs' solution for a minimum of 30 min. Cumulative concentration:response curves to oxytocin (Syntocinon, Sandoz Products Limited, Feltham, London), AII (Sigma Chemical Products, St Louis, USA) or ergometrine (ergometrine maleate; Antigen Ltd, Roserea, Eire) were then established. The final concentrations used were 10^{-11} to 10^{-8} M for oxytocin ($n = 24$ tissues), 10^{-11} to 10^{-5} M for AII ($n = 20$) and 10^{-9} to 10^{-5} M for ergometrine ($n = 24$). These concentrations were achieved by the stepwise addition of stock solution to the bath. Tissues were left in contact with each concentration for 20 min. A recovery period of 40 min followed completion of the initial curves. Tissues were then randomly allocated using a latin square sequence to treatment either with nitrendipine at 10^{-9} M or with its carrier (Odum

& Broughton Pipkin 1987). Tissues were bathed in the Krebs' solution with additive for a minimum of 20 min before the concentration:response curve to the initial agonist was re-established. Experiments were made under sodium light because of a degree of sensitivity to light of nitrendipine.

Contractions were measured as the changes in length from the stable baselines attained during the equilibration periods. The initial response to 100 mM KCl was deemed to be maximal, and contractions were calculated in relation to this for the hormones. EC_{50} s (that concentration of agonist required to evoke a response 50% of maximal) were calculated from logit transformation of the concentration:response curves.

Arithmetic means (SEM) are cited where the data were approximately normally distributed. Two-way ANOVA was used to determine the effects of agonist dose and treatment on response, with Scheffé's test for multiple comparisons.

Results

The myometrial strips came from 24 women (18 first and six second pregnancies) of mean age 27.5 (SE 5.3) years, who were delivered at term by elective lower segment caesarean section under general ($n = 20$) or epidural anaesthetic ($n = 4$). The indications for the elective section were pre-eclampsia ($n = 11$), defined as a persistent systemic arterial pressure at or above 140/90 mmHg with proteinuria in a previously normotensive woman, or non-hypertensive obstetric indications ($n = 13$). No differences were observed in strips from normotensive or hypertensive women. Three women smoked during pregnancy.

All the strips exhibited very minor spontaneous contractile activity which was more or less regular after 60–90 min of equilibration in the Krebs' solution.

Oxytocin as an agonist

The contractile response to oxytocin developed slowly at the lower two concentrations used, but at 10^{-9} and 10^{-8} M contraction developed rapidly and with marked increases in amplitude. The response at 10^{-8} M was close to that elicited by 100 mM KCl. The overall EC_{50} was $8.2 \times 10^{-10} \text{ M}$. As Fig. 1 shows, the mean second concentration:response curve of the control

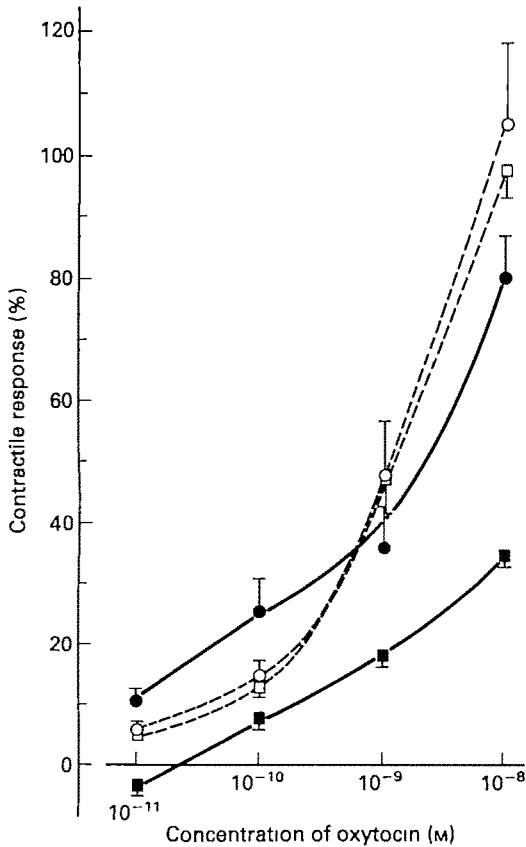


Fig. 1. Concentration:response curves to oxytocin of strips of pregnant human myometrium. Responses are expressed as percentages of each strip's initial response to 100 mM KCl. Curves obtained during the first part of the experiment are shown as $\bigcirc$ ----- $\bigcirc$ and $\square$ ----- $\square$; the second curves were obtained in the presence of carrier ($\bullet$ — $\bullet$) or nitrendipine 10^{-9} M ($\blacksquare$ — $\blacksquare$). Mean values (SEM) are shown, with lines of best fit drawn in by eye. ANOVA revealed a statistically highly significant effect ($P < 0.001$) of nitrendipine on the response to oxytocin, an effect which was most marked at high agonist concentrations

tissues ($\bullet$ — $\bullet$) was very similar to the first ($\bigcirc$ --- $\bigcirc$), from which it did not differ significantly ($P > 0.25$). However, there was blunting of response in the presence of nitrendipine, such that 11 of the 12 tissues actually showed a small relaxation at the lowest concentration of oxytocin. The difference between the curves for control and nitrendipine-treated tissues was highly significant ($P < 0.001$); differences between individual values only reached statistical significance at the highest dose ($P < 0.001$). An EC_{50} could not appropriately be calculated

for the nitrendipine-treated tissues because of the marked blunting of response.

Angiotensin II as an agonist

A much wider concentration range was required for AII than for oxytocin to elicit a near-maximal concentration:response curve (see Figs 1 & 2). While most tissues showed a gradual dose-dependent contraction to AII, in three there was a sudden rapid contraction of considerable amplitude starting at a concentration of 10^{-7} M AII, and resembling that seen in response to the higher doses of oxytocin. These tissues came from three different patients. The initial concentration:response curves were effectively identical in the two treatment groups (Fig. 2)

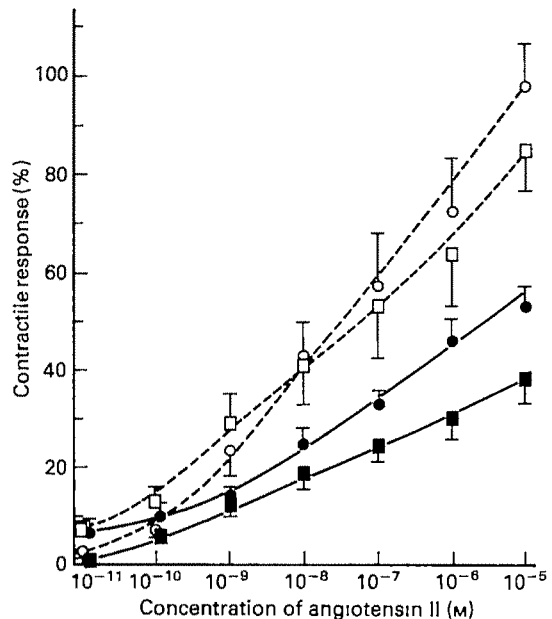


Fig. 2. Concentration:response curves to angiotensin II of strips of pregnant human myometrium. Responses are expressed as percentages of each strip's initial response to 100 mM KCl. Curves obtained during the first part of the experiment are shown as $\bigcirc$ ----- $\bigcirc$ and $\square$ ----- $\square$; the second curves were obtained in the presence of carrier ($\bullet$ — $\bullet$) or nitrendipine 10^{-9} M ($\blacksquare$ — $\blacksquare$). Mean values (SEM) are shown, with lines of best fit drawn in by eye. The contractile response to angiotensin II was diminished in both vehicle- and nitrendipine-treated tissues in the second part of the experiment. This is presumed to be partly due to tachyphylaxis, but ANOVA revealed a statistically highly significant effect ($P < 0.001$) of nitrendipine on the response to angiotensin II, an effect which was most marked at high agonist concentrations.

with an overall EC_{50} of $3.4 \times 10^{-8} M$. However, both treatment groups showed a significant blunting of response during the second part of the experiment, although this was substantially more marked in the tissues treated with nitrendipine (ANOVA, $P < 0.001$). Indeed, only two tissues in this group responded at all to $10^{-11} M$ AII, compared with three non-responders in the control group. Again, the effect of nitrendipine appeared to be more marked at higher concentrations of AII ($10^{-7} M$, $P < 0.05$; 10^{-6} and $10^{-5} M$, $P < 0.001$).

Ergometrine as an agonist

Pilot studies had shown myometrial strips to be unresponsive to ergometrine at concentrations below $10^{-9} M$; above this concentration, however, the response was rapid and sustained, becoming near-maximal at $10^{-5} M$. The initial concentration:response curves were identical in the two treatment groups (Fig. 3), with an overall EC_{50} of $3.9 \times 10^{-8} M$. Both treatment groups

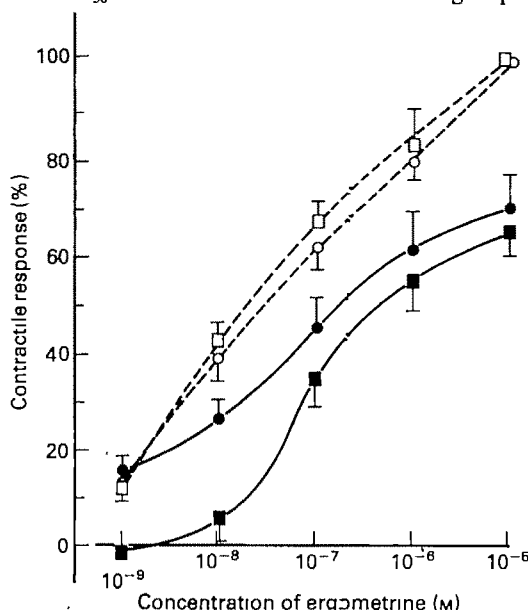


Fig. 3. Concentration:response curves to ergometrine of strips of pregnant human myometrium. Curves obtained during the first part of the experiment are shown as $\bigcirc$ ----- $\bigcirc$ and $\square$ ----- $\square$; the second curves were obtained in the presence of carrier ($\bullet$ — $\bullet$) or nitrendipine $10^{-9} M$ ($\blacksquare$ — $\blacksquare$). Mean values (SEM) are shown, with lines of best fit drawn in by eye. ANOVA revealed a statistically highly significant effect ($P < 0.001$) of nitrendipine on the response to ergometrine, an effect which was most marked at low agonist concentrations.

showed significant overall blunting of response during determination of the second concentration:response curves, but the effect was considerably more marked in the tissues exposed to nitrendipine (ANOVA, $P < 0.001$). Only two tissues in this group responded to ergometrine at $10^{-9} M$, compared with 10 in the control group, and five still failed to contract at $10^{-8} M$. These differences at the lower doses were statistically significant ($P < 0.05$ for both) but were not significant at higher doses.

Discussion

The concentration range of oxytocin studied covered effectively all of the tissue response curve, with the concentration for maximal contraction closely resembling that described by Maigaard *et al.* (1986b). However, Maigaard *et al.* (1986b) demonstrated only a moderate inhibition of response to their standard test concentration of oxytocin $10^{-8} M$ by nitrendipine $10^{-9} M$, while we found a marked response (Fig. 1). This difference is probably one of methodology related to the longer time of exposure to agonists of the tissues in this study. At the lower concentrations of oxytocin studied in this paper nitrendipine was without significant effect on the response. This would be in agreement with the enhancing effect of oxytocin on the release of intracellularly bound calcium ions (Carsten 1979) to initiate contraction rather than an action by increasing the influx of extracellular calcium ions. It seems possible that this second mechanism comes into play at high oxytocin concentrations, such as could be encountered during labour. Incubation of myometrial strips in calcium-depleted medium blocked the response to oxytocin $10^{-8} M$ (Maigaard *et al.* 1986b) which supports this hypothesis.

AII is the most potent naturally occurring circulating vasoconstrictor known, but as Fig. 2. shows, its effects on pregnant myometrium are much less. The wide span of the concentration:response curve suggests that it is unlikely to play a physiological role as an oxytocic agent. Even at delivery, plasma concentrations of AII in excess of $10^{-10} M$ are rarely encountered (Broughton Pipkin & Symonds 1977), while the initial EC_{50} here was $3.4 \times 10^{-8} M$. It is, however, possible that there might be locally high concentrations of AII as a consequence of production by the myometrium itself. Pregnant myometrium can synthesize

renin in tissue culture (Symonds *et al.* 1968) and renin-like granules have been shown in uterine muscle (Johnson *et al.* 1984).

The rightward displacement and blunting of the second control concentration response curve to AII is presumably due to tachyphylaxis, as previously described in this preparation (Maigaard *et al.* 1986b). Treatment with nitrendipine further blunted the response at high AII concentrations, in a manner similar to that seen with oxytocin. AII is known to exert its primary contractile effect on vascular smooth muscle by mobilizing calcium from intracellular storage sites (Alexander *et al.* 1985) but its sustained contractile effects require the influx of extracellular calcium. These two factors together can explain the observed effect of nitrendipine (Fig. 2).

The ergot derivative ergometrine is used in the treatment of postpartum haemorrhage, and its constrictor effect is mainly confined to the uterus. Tachyphylaxis is presumably again the explanation for the blunting of response to ergometrine seen in the second control concentration:response curve. Myometrial tissue strips do show rapid tachyphylaxis *in vitro* to a variety of natural contractile agents (Maigaard *et al.* 1986c). If such tachyphylaxis occurs *in vivo* it might contribute to the length of labour. The inhibition by nitrendipine of ergometrine-induced contraction at low concentrations (Fig. 3) might be of concern if nitrendipine were to be used in the treatment of hypertension in pregnancy. However, the use of ergometrine is in any case contraindicated in hypertensive pregnancy because of its constrictor effect on the arterioles.

Myometrial contractility is heavily dependent on the availability of extracellular calcium (Bolton 1979; Forman *et al.* 1986) so that agents, which inhibit the flow of calcium across the cell membrane would be expected to inhibit uterine contractility. Such, indeed, is the case (e.g. Csapo *et al.* 1982; Sakamoto & Huszar 1984; Maigaard *et al.* 1986a). The data presented here show that nitrendipine also inhibits the myometrial response to both oxytocin and ergometrine, both of which exert powerful oxytocic effects on the uterus through an effect on calcium flux. The possible clinical implications are obvious.

Preliminary data from animal and human studies suggest that dihydropyridines like nitrendipine are indeed at least as effective as β -agonists in suppressing preterm labour, and may in

fact be of greater efficacy (Sakamoto & Huszar 1984; Golichowski *et al.* 1985; Read & Wellby 1986). They are also reported to have a lesser effect on maternal heart rate and other indices of cardiac function, although some fetal tachycardia has been reported (Golichowski *et al.* 1985; Read & Wellby 1986). Their clinical use would be logical, in that it blocked part of the contractile mechanism of the myometrium. Further direct investigation of their clinical usefulness as tocolytic agents seems warranted.

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Intrauterine pressure studies in multigravid patients in spontaneous labour: effect of oxytocin augmentation in delayed first stage

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Summary. Uterine activity was assessed by measuring the uterine activity integral (UAI) in 116 black multigravid patients in spontaneous active labour at term. Although individual patients showed a wide range of mean UAI levels, normal labour (group I, $n = 54$) was associated with a mean UAI level of 1640 kPas/15 min which was higher than the mean level observed in patients in whom delay in cervical dilatation occurred in active labour. The patients who failed to progress in labour were treated with oxytocin infusion and 32 of them made good progress in labour and achieved vaginal delivery (group IIa: mean UAI pre-oxytocin treatment 1040 (SD 424) kPas/15 min, post-oxytocin 1890 (SD 559) kPas/15 min). The other 23 patients required operative delivery (group IIb) despite correction of uterine activity after oxytocin treatment (pre-oxytocin mean UAI 1230 (SD 570) kPas/15 min, post-oxytocin 1815 (SD 650) kPas/15 min). The rate of oxytocin infusion varied between patients from 2 to 16 mU/min but in 75% uterine activity was corrected to normal levels by a dose of ≤ 8 mU/min. It is concluded that delay in progress in labour is associated with lower intrauterine pressures than are present in normal labour, and that management of patients presenting with inefficient uterine action may benefit from the use of uterine activity integral measurements.

The importance of assessing uterine activity in the clinical monitoring of the progress of labour is well established (Gordon 1984; Steer *et al.*

1984; Arulkumaran *et al.* 1984). A common cause for prolonged labour is inefficient uterine action for which the usual management is to rupture the membranes and commence an intravenous infusion of oxytocin to promote uterine activity once secondary causes such as cephalopelvic disproportion and malpresentation have been excluded.

Objective determination of the correct dose of oxytocin infusion, however, is difficult. While frequency and duration of contractions are easily determined clinically, 'strength' (active pressure, which is an important factor in determining cervical dilatation) is much more difficult to assess.

Accurate assessment of the strength of uterine

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contractions is now possible with the measurement of the uterine activity integral (UAI) or active contraction area (Steer *et al.* 1984). This measurement incorporates the variables of frequency, duration and strength of contractions and has shown a higher correlation with the rate of progress of labour than the Montevideo unit which does not include the effect of duration of contractions (Steer *et al.* 1984).

Using UAI measurements, recent studies have reported on levels of uterine activity in spontaneous labour. These reports, however, vary greatly in the mean UAI levels obtained in normal labour, both in primigravid and multigravid patients (Steer *et al.* 1984; Arulkumaran *et al.* 1984). Furthermore, there have been conflicting data on UAI measurements in multigravid patients in comparison to their primigravid counterparts. While Arulkumaran *et al.* (1984) reported lower UAI levels in multigravid patients, Steer *et al.* (1984) found the converse to be true.

Using the apparatus described by Steer *et al.* (1984) we have studied the range of UAI levels during normal labour in local black multigravid patients. In multigravid patients requiring oxytocin augmentation for delay in progress in

labour we also studied the dose of oxytocin required to effect normal vaginal delivery in relation to the UAI measurements following oxytocin administration.

Patients and methods

All patients included in the study were black multigravidae at term, with a history of previous normal (unassisted) vaginal deliveries and presenting in the active phase of spontaneous labour (cervix fully effaced and at least 3 cm dilated with painful uterine contractions). The study was undertaken over a period of 20 months.

Written informed consent was obtained from all patients before they were transferred to an intensive monitoring area of the labour ward where each patient was personally attended by the same midwife specially trained in the techniques of the study. The University of Natal ethics committee gave approval for the study.

Intrauterine pressures were measured using the apparatus and method as described in detail by Cowan *et al.* (1982a). This apparatus integrates the area under each intrauterine pressure curve above the resting tone of the uterus, summates the area under successive pressure curves

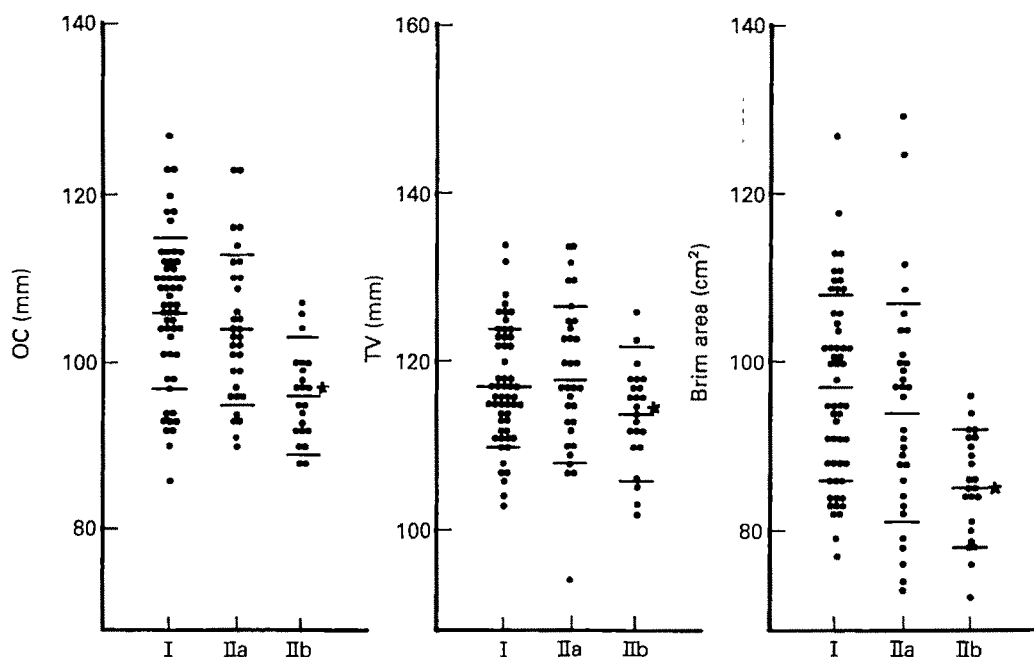


Fig. 1. X-ray pelvimetry measurements (obstetric conjugate: OC; transverse diameter: TV; and brim area) in three groups of patients (group I, normal labour, group IIa, delayed labour, vaginal delivery, group IIb, delayed labour, operative delivery). All X-rays were performed within 24 h of delivery. Horizontal bars indicate the mean $\pm$ SD
* $P < 0.05$ compared with groups I and IIa.

and displays the summated integral (in kPas) every 15 min.

After an initial clinical assessment a Portex intrauterine catheter (700/415/010) attached to an external pressure transducer (Bell & Howell type 4-327-1) was introduced through the cervical canal until the tip of the catheter was placed beyond the fetal head. The catheter was flushed with sodium chloride solution (150 mmol/l saline) by gravity feed during the introduction into the uterine cavity and repeated briefly after each new display reading on the integrator to ensure patency of the intrauterine catheter.

Uterine activity was measured until the cervix became fully dilated. The second stage of labour is not included in the present study. Fetal condition was continuously monitored by cardiotocography using a scalp electrode and fetal blood pH estimation where necessary.

Progress in labour was assessed hourly using an aseptic vaginal technique. Details of labour were recorded on a special datasheet as well as on a partogram (Philpott 1972). Gestational age at delivery was assessed as described by

Dubowitz (1970). The biparietal diameter was measured by the same person 12 h and 5 days after delivery using engineering calipers and a mean of six readings was taken. X-ray pelvimetry was performed on all mothers within 24 h of delivery.

Uterine activity was analysed in two ways. First, all the recorded values in each group (see results) were pooled and analysed collectively in relation to cervical dilatation. In those patients requiring oxytocin augmentation the mean uterine activity for each patient was assessed both before and after oxytocin administration. Second, uterine activity was also analysed individually for each patient so that the distribution of the mean level could be compared between groups.

Statistical analysis

The unpaired *t*-test was used to assess the differences in the clinical characteristics of mothers and babies in groups I and II. The position of the fetal head was compared between groups using

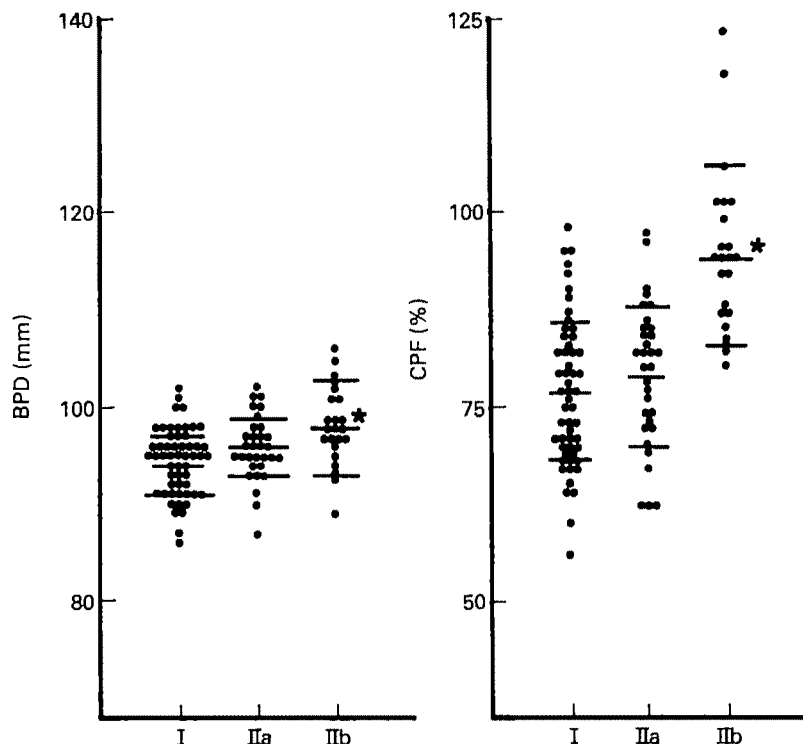


Fig. 2. The biparietal diameter (BPD) and cephalopelvic fit (CPF) measurements of babies born to the three groups of patients (group I, normal labour; group IIa, delayed labour, vaginal delivery; group IIb, delayed labour, operative delivery). Horizontal bars indicate the mean $\pm$ SD * $P < 0.05$ compared with group I.

the χ^2 -test. The Mann-Whitney *U*-test was applied to the intrauterine pressure data. A significance level of 5% was chosen for all tests.

Results

A total of 116 patients were included in the present study. They were categorized retrospectively into four groups depending on the outcome of labour.

Group I: normal labour

The 54 patients in this group presented in spontaneous labour, progressed at a cervical dilatation rate of >1 cm/h, and were delivered vaginally.

Group II: delayed labour

The 55 patients in this group were all admitted in spontaneous labour but failed to progress adequately in the absence of obvious clinical signs of cephalopelvic disproportion. These patients were treated with oxytocin infusion (2 mU/min increased arithmetically, maximum infusion rate being 16 mU/min). This group was further subdivided depending on the outcome of labour following oxytocin administration.

Group IIa. The 32 patients categorized to this subgroup made good progress after oxytocin (cervical dilatation >1 cm/h) and were delivered vaginally without assistance.

Group IIb. This group comprised the other 23 patients who failed to progress adequately after oxytocin administration and required operative

delivery. Two of these patients required operative delivery because fetal distress was diagnosed as evidenced by late decelerations on the fetal heart monitor and decreased fetal scalp pH measurements.

Group III

There were seven patients who presented with delay in active labour and were not treated with oxytocin; four developed cervical arrest at 9–10 cm cervical dilatation and required instrumental vaginal delivery and the other three had cervical arrest at 6–8 cm dilatation despite strong uterine contractions, and were delivered by caesarean section.

There were no statistical differences in maternal age or the weight of the babies between all four groups of patients. The pelvic diameter measurements were not statistically different between groups I and IIa (Fig. 1). Although the biparietal diameter (BPD) of babies in group IIa was marginally greater than that in group I ($P = 0.05$) there was no statistically significant difference in the cephalopelvic fit (CPF) between the two groups (CPF is the relation between the head area of the baby and the brim area of the pelvis and is expressed as a percentage) (Fig. 2). However, the women in the group requiring operative delivery (group IIb) had significantly lower obstetric conjugate (OC) and transverse diameter (TV) measurements than the women in groups I and IIa ($P < 0.05$) (Fig. 1). Also, the mean BPD was increased in these babies resulting in a larger CPF measurement

Table 1. Mean intrauterine pressure recordings in the various groups of multigravid patients

Patient group	No. of patients	Uterine activity integral (kPas/15 min) Mean (SD)	No. of recordings
Normal labour (I)	54	1640 (690)	319
Delayed labour			
Vaginal delivery (IIa)			
Before oxytocin	32	1040* (424)	176
After oxytocin	32	1890** (559)	197
Operative delivery (IIb)			
Before oxytocin	23	1230* (570)	133
After oxytocin	23	1815** (650)	197
No oxytocin (III)	7	1750 (490)	70

* $P < 0.002$ compared with group I.

** $P < 0.001$ compared with corresponding mean levels before oxytocin treatment

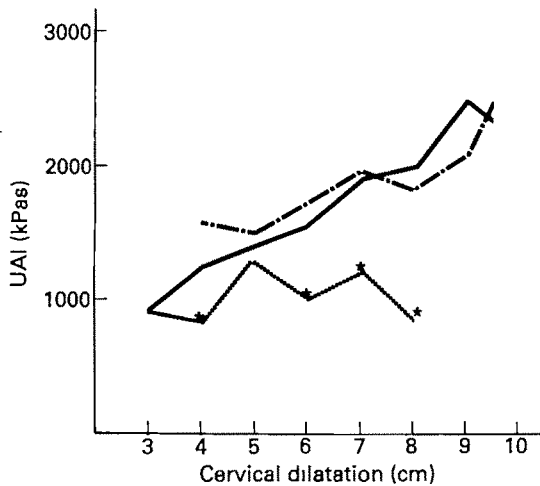


Fig. 3. Relation of uterine activity (UAI) to cervical dilatation in normal labour (—) and delayed labour (group IIa) before oxytocin (.....) and after oxytocin (-.-.-). Significantly lower UAI levels were obtained at 4, 6, 7 and 8 cm cervical dilatation before oxytocin treatment in delayed labour. * $P < 0.001$

than in groups I and IIa ($P < 0.05$) for both (Fig. 2).

Analysis of the positions of the fetal head at the time of arrest of cervical dilatation in group IIa showed no statistically significant difference from those found in patients in normal labour on admission. Occipito posterior position was present in only 3% of patients who developed cervical arrest and in 2% of those who had normal labours.

Analysis of uterine activity

Collective analysis. The mean intrauterine pressure measured by the uterine activity integral (UAI) of all recordings in each group is presented in Table 1. Normal labour was associated with a mean UAI of 1640 (SD 690) kPas/15 min. In patients requiring augmentation of labour the mean UAI before oxytocin was significantly less than in normal labour. In the seven patients not treated with oxytocin the mean UAI was similar to that in normal labour. Figure 3 demonstrates the relation between UAI levels and cervical dilatation. Normal labour is associated with a significant increase in UAI as labour progresses ($P < 0.05$). In patients who were delivered vaginally after oxytocin administration, UAI levels at 4, 6, 7 and 8 cm cervical dilatation before oxytocin treatment were significantly lower than the corresponding values in normal labour

($P < 0.001$) (Fig. 3). Mean UAI values after oxytocin administration in this group were not significantly different from those in normal labour at any stage of cervical dilatation (Fig. 3). The dose of oxytocin required to correct the uterine activity level varied between patients (Table 2). Although the maximum infusion rate was 16 mU/min, 75% of patients in group IIa were delivered vaginally after a maximal dose of ≤ 8 mU/min.

Individual analysis. Figure 4 shows the distribution of mean levels of UAI in all patients in group I and in group II (before and after oxytocin administration). Clearly depicted is the wide range of uterine activity in normal labour. Only five patients presenting with delayed labour (four in group IIa and one in group IIb) had mean UAI levels greater than the mean level found in the group with normal labour (Fig. 4). All patients in group II(a+b) demonstrated an increase in mean UAI after oxytocin treatment. The reasons for operative delivery in group IIb were fetal distress in two patients (mean UAI levels after oxytocin treatment: 2350 and 1760 kPas/15 min respectively) and failure to progress following oxytocin augmentation.

Morbidity and mortality

All infants were in good condition at birth, all had Apgar scores > 7 at 5 min and an uneventful neonatal progress.

Discussion

Normal labour has arbitrarily been defined in the present study as labour of spontaneous onset progressing at a cervical dilatation rate > 1 cm/h in the active phase. This was based on the finding by Philpott & Castle (1972) that the slowest 10% of primigravid Africans in spontaneous active

Table 2. Maximum dose of oxytocin administered to patients in group II

Oxytocin dose (mU/min)	No. of patients	
	Group IIa	Group IIb
2	1	2
4	4	5
6	5	3
8	14	9
10	6	2
12	2	1
14	—	1

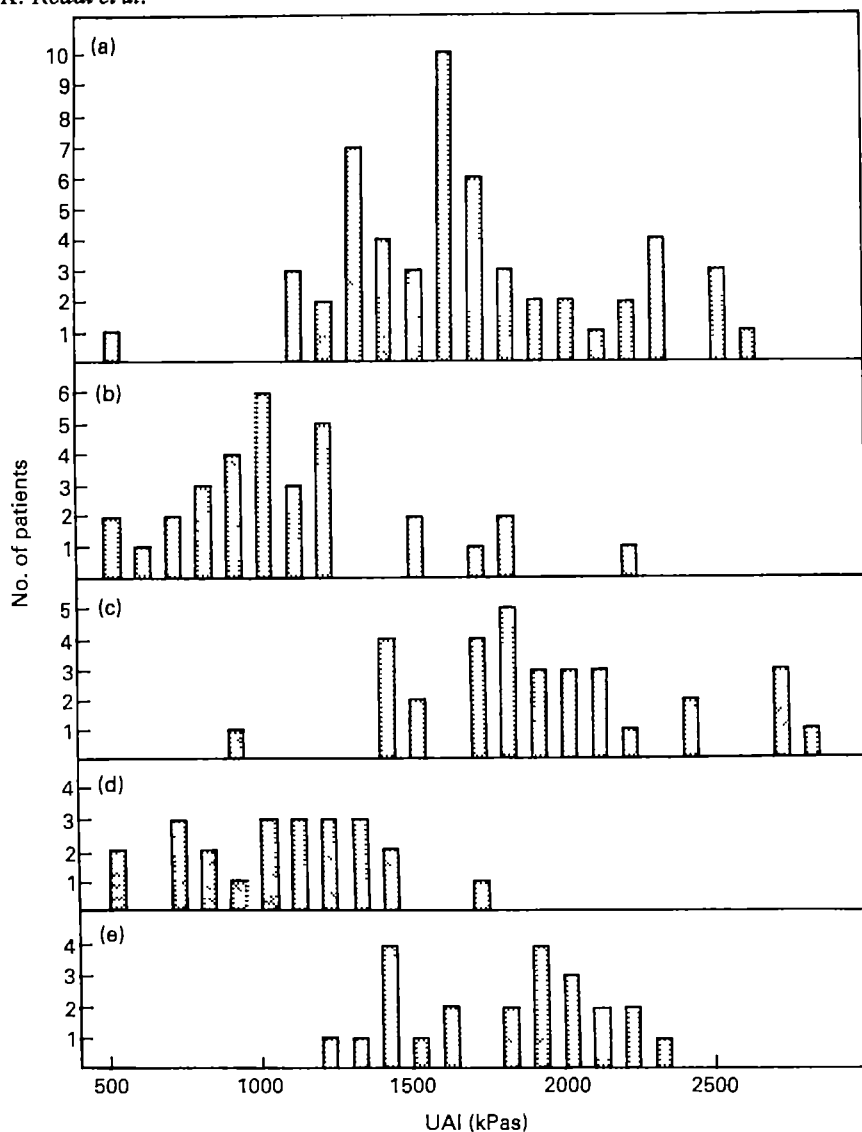


Fig. 4. Distribution of mean levels of uterine activity (UAI) in patients in different groups of multigravid patients before and after oxytocin treatment. (a) Normal labour; (b) delayed labour, group IIa, pre-oxytocin; (c) delayed labour, group IIa, post-oxytocin; (d) delayed labour, group IIb, pre-oxytocin; (e) delayed labour, group IIb, post-oxytocin.

labour had a mean cervical dilatation rate of 1 cm/h. Since multigravid patients seem to have more rapid labours, a higher percentage would be expected to progress at a cervical dilatation rate of over 1 cm/h. Our choice of a line drawn at 1 cm/h in the active phase of labour to distinguish slow from normal labour is therefore conservative.

The mean UAI in normal labour in the present series of 1640 kPas/15 min is slightly less than that reported by Cowan *et al.* (1982a,b) who studied primigravid patients in the same popula-

tion group, but higher than the average UAI reported in spontaneous labour by other groups (Steer *et al.* 1984; Arulkumaran *et al.* 1984). It is possible that the common occurrence of pelvic contracture in the local black population contributes considerably to pelvic resistance in these patients and that greater uterine forces are generated to effect vaginal delivery. Also, admission of patients late in active labour into the study may result in a higher mean UAI level (50% of patients in the present study were admitted at cervical dilatation ≥ 5 cm).

Of interest in the present study is the consistently lower mean UAI levels in patients who subsequently showed delay in active labour (91% of these patients had exhibited lower mean UAI levels than the mean level in normal labour). However, no distinction could be made between those patients who eventually were delivered vaginally after oxytocin treatment and those who required operative delivery, since both subgroups exhibited similarly lower UAI levels during the pre-oxytocin period compared with the normal labour group.

Once uterine activity was corrected to correspond with levels in normal labour, these two subgroups differed in the clinical response with respect to cervical dilatation and progress in labour. The presence of marginal cephalopelvic disproportion in group IIb is probably the cause of the failure to progress after oxytocin administration. Whether the poor uterine activity in this group was secondary to cephalopelvic disproportion is uncertain. UAI measurements in multigravid patients with obvious clinical signs of this condition have not been assessed and this might be helpful in establishing the response of the gravid uterus to cephalopelvic disproportion.

Although the rate of oxytocin infusion varied between 2 mU/min and 16 mU/min in the present series, most patients were delivered on infusion rates of ≤ 8 mU/min. This is considerably less than the infusion rates reported by Cowan *et al.* (1982b) in the primigravid patients. It was also noted that in 10 patients (30%) in group IIa oxytocin infusion rates needed to be decreased once effective uterine contractions were obtained. Oxytocin infusion rates were decreased in three patients (13%) in group IIb.

The present study confirms the importance of uterine activity measurements in the investigation of failure of the cervix to dilate in spontaneous labour. In the absence of a clinical response following correction of uterine activity, operative delivery should be considered. It is emphasized also that intensive monitoring of these patients during oxytocin infusion be undertaken so that hyperstimulation may be recognized early and infusion rates decreased if necessary.

Finally, in a previous study on prostaglandin (PG) concentrations in amniotic fluid in multigravid patients presenting with delayed labour in the absence of cephalopelvic disproportion, sig-

nificantly lower PGF concentrations were observed compared to normal labour (Reddi *et al.* 1984). Since PGF concentrations did not increase after oxytocin treatment and correction of uterine activity in these patients, there exists the possibility of an intrinsic biochemical defect in PG production within the feto-placental unit in patients presenting with primary inefficient uterine action (Reddi *et al.* 1987).

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Multiple caesarean sections: outcomes and complications

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Summary. The outcome and complications of caesarean section were compared between 64 women who had had between three and eight previous sections (mean four) and 61 women sectioned for the first or second time. Gestational age was slightly less in the group with multiple operations. Of the women with three or more previous sections 27% had developed a fenestration in the transverse uterine scar; this was symptomless in half of them, and there was no associated severe ante- or intrapartum haemorrhage. Operation time was longer in women with multiple caesarean sections, and 13% required a caesarean hysterectomy. There were no other differences in the operative or postoperative courses between the two groups. In most women who have had multiple caesarean sections it is possible to wait for elective delivery until the fetus is mature. No absolute upper limit for the number of repeat caesarean sections can be given.

It is generally assumed that the improvements in the safety of anaesthesia, pre- and postoperative monitoring, antibiotics and the availability of blood products, together with altered obstetric practices, are the reasons which have increased the caesarean section rate all over the world. Furthermore, a previous caesarean section has acquired increasing importance as an indication for delivery by caesarean section in the next pregnancy (Rosen *et al.* 1981; Yudkin & Redman 1986). Although we have a great deal of experience in trials of labour after a previous caesarean section (Lavin *et al.* 1982; Martin *et al.* 1983; Flamm 1985), it is still considered a complicating factor in the obstetric care of later pregnancies and deliveries. Modern medicine has facilitated repeated operations, but little is known about the risks involved in multiple caesarean sections.

This report describes the operative course and outcome of pregnancies in women who have had several previous caesarean sections (range 3-8).

Patients and methods

The study group included 64 pregnant women, who had had three or more previous caesarean sections, managed by the Department of Obstetrics and Gynecology, University of Oulu, Finland, between 1978 and 1986 (inclusive). Most of these women belong to a religious group whose members do not accept contraception or sterilization. One had had eight previous sections, two had had six, 12 had had five, 14 had had four and 35 had had three previous caesarean sections. Elective caesarean section was the planned method of delivery for each of these women and in the event 45 (70%) were sectioned before the onset of labour or rupture of the membranes and the other 19 women were sectioned in early labour within 2 h of the onset of the contractions. A control group was formed by selecting the next woman delivered by caesarean section after each of the 64 women in the study group. Three women in the control group

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were excluded from the final analysis because of missing data, so that the final control/comparison group, used to represent a series of typical caesarean sections, comprised 61 patients. Nine women in the control series had had one previous section and one had had two. All previous sections in both groups were made using a low transverse uterine incision. All these patients had had regular antenatal care. Antenatal admission for longer than 1 week was needed in 64% of the study group and in 16% of the control group. Of the 61 women in the control group 23 (38%) had elective sections, and 38 (62%) were sectioned in labour.

The clinical features, indications for the primary section and the operative details are presented in Table 1. The operations were performed by members of the same staff of the clinic (in all cases senior obstetricians or well-trained

assistant doctors). Either Pfannenstiel or midline longitudinal abdominal incisions were used, and the standard transversal lower segment incision was made in the uterus whenever possible. Early mobilization (within 24 h) was practised, except for two patients who needed post-operative intensive care.

Comparison between the operative and post-operative courses and perinatal data between the groups was made by the Mann-Whitney *U*-test (continuous variables) and the χ^2 test (discrete variables). A *P*-value less than 0.01 was used for the level of significance.

Results

There were no maternal deaths. The perinatal data are presented in Table 2; there were no significant differences between the two groups.

Fenestrations

In the study group, uterine scar fenestration (fetal membranes visible at a uterine window before incision) was seen in 17 women (27%) (Fig. 1). The prevalence of fenestration was significantly higher in women who had had more than three sections (38%) than in those who had had only three previous sections (17%). Eight (47%) of the fenestrations were clinically silent and not suspected before the operation. One woman with a silent fenestration had had slight vaginal bleeding during the last month of gestation, and seven had felt pain in the region of the old scar. No severe antenatal haemorrhage occurred in any woman. The prevalence of fenestration did not differ between women sectioned before labour and those sectioned in labour. None of the women in the control group had a uterine scar fenestration.

Operative aspects and complications

The duration of the operation, calculated from the point when anaesthesia (either general or local) allowed the skin incision to be made to the last skin suture, and other operative data are presented in Table 3.

The baby was born (timed from the beginning of the skin incision to clamping of the cord) approximately 4 min earlier in the control group than in the study group. This difference did not affect the Apgar scores. With general anaesthesia the mean time to birth was 4.3 (SD 2.5)

Table 1. Clinical features and indication for primary caesarean section (CS) in the two groups

Variable	Study group (multiple CS) (<i>n</i> = 64)	Control group (<i>n</i> = 61)
Maternal age (years)		
mean (SD)	32.1 (5.1)	26.8 (5.1)*
Parity		
median (range)	4 (3–12)	1 (0–5)*
Years since first pregnancy		
median (range)	8 (4–20)	4 (2–9)*†
Years since last pregnancy		
median (range)	2 (1–15)	2 (1–4)†
Indication for primary CS	<i>n</i> (%)	<i>n</i> (%)
Fetopelvic		
disproportion	51 (80)	22 (36)*
Other‡	8 (13)	30 (49)*
Maternal disease	5 (8)	9 (15)
Mode of operation		
Elective	45 (70)	23 (38)*
Non-elective	19 (30)	38 (62)*
Anaesthesia		
Spinal/epidural	48 (75)	38 (62)
General	16 (25)	23 (38)
Mode of incision		
Pfannenstiel	23 (36)	47 (77)*
Midline	41 (64)	14 (27)*
Gestational age at delivery (weeks)		
mean (SD)	38.0 (2.2)	39.2 (2.3)*

Significance of difference between the two groups
* *P* < 0.01.

† Nulliparas are excluded

‡ Other obstetric indications include prolonged labour, breech, fetal distress, etc

min in the study group and 3.5 (SD 2.3) min in the control group. With spinal or epidural anaesthesia, where there is more time available for safe wound preparation, the corresponding mean times were 10.2 (SD 6.9) and 7.5 (SD 4.2) min. Four women (6%) in the study group had a large rectal muscle diastasis on both sides of the old longitudinal scar and 39 (60%) had fused abdominal wall layers with dense scar tissue. The urinary bladder was attached high on the anterior wall of the abdomen in 24 (37%) women but intraperitoneal adhesions were found in only seven. The urinary bladder was entered accidentally in four women and treated by primary suture. In seven women with fenestrations, difficulties in repairing the severely lacerated or membranous margins of the uterine wound necessitated an immediate subtotal hysterectomy. In one woman atonic bleeding made emergency hysterectomy necessary. Postoperative atonic bleeding (more than 1000 ml within the first 4 h) occurred in three patients. In one, a large resection of omentum was needed because of damage caused while extracting the baby.

In the control series, two patients had intraperitoneal adhesions and five had thickened and fused abdominal wall layers in their old scar. Two deep cervical ruptures, caused by the extraction of deep-lying fetal heads, occurred in the control series, and were successfully sutured. Four control patients had postoperative atonic bleeding, treated conservatively. Prophylactic antibiotic therapy was initiated at the operation in four patients in the study series (urinary blad-

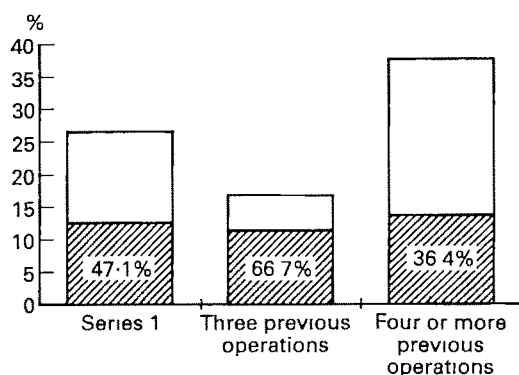


Fig. 1. The prevalence of lower segment fenestrations in 64 women with multiple caesarean sections. The shadowed area represents the proportions of patients without clinical symptoms. Series 1 = the whole study group.

Table 2. Perinatal features in the two groups

Variable	Study group (n = 64)	Control group (n = 61)
Birthweight (g)		
mean (SD)	3207 (556)	3370 (754)
Apgar score		
median (range)		
At 1 min	9 (4-10)	9 (0-10)
At 5 min	9 (3-10)	9 (0-10)
No. below 10th		
birthweight centile	2	6
No. of preterm births		
(<36 weeks)	7	5
No. of twin pregnancies	0	4
No. of perinatal deaths	1*	2†

* Severe rhesus isoimmunization.

† Placental abruption.

der ruptures) and in 12 of the control series (prolonged labour).

Postoperative course

Details of the postoperative complications and the length of the postoperative care are presented in Table 3. There were no significant differences between the two groups. Intensive postoperative care was needed in one patient with severe pre-eclampsia in the control series, and in one in the control group both ante- and postnatally for tuberculous meningitis. No clinically evident thromboembolic complications or sepsis occurred, nor paralytic or mechanic ileus. In the study group the postoperative complication rates did not differ between the women who were sectioned before the start of labour and those sectioned in labour, but in the control group those sectioned in labour had postoperative fever more often and more blood loss at the operation. Those having caesarean hysterectomy had more blood transfusions and febrile postoperative days than the other patients, but the length of the postoperative care was the same.

Discussion

The two previous reports involving many women with multiple caesarean sections date from the 1960s. In one (Piver & Johnston 1969) the outcome and complications were comparable to those in the present report, but the other (Browne & Hynes 1965) reported an increased

Table 3. Operative and postoperative course of the patients in the two groups

Variable	Study group (<i>n</i> = 64)	Control group (<i>n</i> = 61)
Duration of operation (min)	63.8 (22.4)	46.9 (18.5)*
Incision to delivery interval (min)	9.2 (6.6)	5.0 (3.6)*
Blood loss at operation (ml)	681 (661)	789 (489)
Haemoglobin decrease (g/l)	10.8 (14.1)	12.1 (12.5)
Days of postoperative care	8.9 (1.2)	9.7 (1.3)
Caesarean hysterectomies	8 (13%)	0**
Injuries at operation		
Bladder injury	4 (6%)	0
Deep cervical rupture	0	2 (3%)
Postoperative course		
Wound infection	1 (2%)	2 (3%)
Other wound healing problems	3 (5%)	1 (2%)
Endometritis	0	3 (5%)
Urinary tract infection	6 (9%)	1 (2%)
Fever >37.5°C, cause unknown (repeated finding)	15 (23%)	18 (30%)
Blood transfusion required	11 (17%)	10 (16%)

Results are mean (SD) or *n* (%) as appropriate.

Significance of difference between the two groups

* $P < 0.01$, ** $P = 0.013$

risk of placenta praevia, bladder adhesions, haemorrhage and neonatal death. Individual case histories of women having several caesarean sections (up to 13) have been published (Potter 1967; Piver & Johnston 1969).

The present study revealed uterine scar fenestration in 27% of women who have had three or more previous transverse lower-segment caesarean sections. This rate is considerably higher than that reported after one previous section (<1%) (Case *et al.* 1971). Clinical signs, such as scar pain or antepartum bleeding, occurred only in half the women, and the antepartum risk of acute rupture with haemorrhage is small. With proper ante- and postnatal care these fenestrations do not seriously threaten the fetus or increase the maternal complications except for the additional difficulties in the proper closure of the uterine wound. None of the women in the study group had a multiple

pregnancy or polyhydramnios and these conditions may involve different risks. Our study provides no data on outcome if labour contractions are allowed to continue for over 2 h. With careful selection and proper management vaginal birth is possible after a previous caesarean section in 60–80% of patients (Flamm 1985). Some experience has been reported of trials of vaginal labour in patients with more than one previous section (Lavin *et al.* 1982; Martin *et al.* 1983; Porreco & Meier 1983; Farmakides *et al.* 1987). Only about 150 such patients have been reported and most of them had had not more than two previous sections. At present, the success and risks of trials of labour in women with several previous caesarean sections are not properly defined. Most obstetricians do not attempt trial of labour after three or more previous sections, especially if the woman has had no previous vaginal delivery. The present finding of impaired lower segment healing would seem to discourage attempting vaginal delivery in such patients, even though the original indication for a caesarean section was no longer relevant.

From the operative point of view, a high number of previous sections is associated with technical difficulty during the dissection of the abdominal wall and the separation of the bladder from the lower segment. These are, however, of minor clinical importance, as the Apgar scores of the newborn and maternal recovery were good and the primary healing of the wound and urinary bladder were uncomplicated. High attachment of the bladder on the abdominal wall is frequent in these patients and must be taken into consideration when opening the peritoneum. Utero-intestinal adhesions are not very common after caesarean sections.

The relative risk of ordinary caesarean section compared with vaginal delivery is 2–11 times greater for maternal mortality and 2–10 times greater for maternal morbidity (Petitti 1985). The most common postoperative complications are endometritis, urinary tract infections and problems of wound healing. The most important indication for an emergency caesarean hysterectomy is a massive haemorrhage, often due to a placenta accreta or a fresh uterine rupture, and postoperative morbidity is 35–60% in these patients (Plauche 1986; Sturdee & Rushton 1986). The present study did not reveal any increase in the postoperative complications in the patients with multiple previous caesarean sections compared with the control series or with

the general complication rates in other published studies. The patients who had subtotal caesarean hysterectomies had as quick a post-operative recovery as the others. This might be due to the fact that no severe bleeding preceded these operations and in most the indication for hysterectomy was prophylactic rather than absolutely essential. Thus the average primary recovery of a woman who has had several previous caesarean sections does not seem to be different from that after an ordinary caesarean section.

A connection between elective repeated caesarean section and persistent neonatal pulmonary hypertension has been reported (Heritage & Cunningham 1985). This could not be established in the present series. If adequate antenatal care is provided and immediate operation is available, it is possible to wait for as complete maturation of the fetus as possible. There was no difference in the condition of the newborn or in the course of the operation between patients sectioned before and after the start of labour.

The women in the present study had considerably more previous caesarean sections than is usual because they belonged to a religious minority whose ethical attitudes towards sterilization and contraception lead to multiparity. It is probable that multiple caesarean sections will also become more common in ordinary communities if the rate of caesarean section continues to increase. On average, the maternal and fetal outcomes in women who have had multiple previous sections do not differ from those in women after an ordinary caesarean section. Thus, if the woman is ready to accept the general risks of an operative delivery and frequently repeated pregnancies, a high number of previous caesarean sections as such does not justify a strong recommendation for sterilization.

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Energy supplementation in the last trimester of pregnancy in East Java: I. Effect on birthweight

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Summary. The effect of two levels of energy supplementation in the last trimester of pregnancy on birthweight was tested in a controlled randomized trial in three villages in Madura, East Java. The high and low energy supplements provided 1.95 MJ (465 kcal) and 218 kJ (52 kcal) per day respectively. In the baseline period the home diet provided on average 6.28 MJ (1500 kcal) (SD 2.1 MJ (499 kcal)) and 41 g (SD 13 g) of protein. The mean birthweight was 2835 g and the rate of low birthweight 12.2%. In the experimental period the home diet was better. The average intake ranged from 6.45 to 7.19 MJ (1541-1717 kcal) and 41.4-44.2 g per day, depending on the degree of compliance. Mean birthweight increased by 100 g and the rate of low birthweight dropped to 9.5%. There was no difference between the high and low energy supplemented group as a whole, probably due to the masking effect of the better home diet in the experimental period. It is likely that a positive effect of energy supplementation on birthweight was restricted to the group of pregnant women with the lowest home dietary intake and/or a low prepregnant weight. In this community targeting of supplementation to lean seasons and/or to women with a low prepregnant weight may be cost-effective.

The high prevalence of low birthweight (LBW) in developing countries indicates that during intrauterine life the fetus is not completely protected from harmful environmental influences (Sterky & Mellander 1978; WHO 1980). Low dietary intakes during pregnancy, notably of energy, may be an important contributing factor (Bergner & Susser 1970; Lechtig *et al.* 1979; Prentice 1980; Monte & Lechtig 1987), but

several authors question the view that energy intake or energy reserves during pregnancy influence fetal growth rate (Grundy *et al.* 1978; McFayden *et al.* 1984; Briend 1985; Durnin *et al.* 1985). The ultimate proof of a direct relation between maternal nutrition and birthweight can only be provided by experimental studies. The results from feeding trials in Guatemala (Lechtig *et al.* 1975a), Colombia (Mora *et al.* 1979), Taiwan (Adair & Pollitt 1985), The Gambia (Prentice *et al.* 1983) and from routine feeding programmes in the USA (Rush 1983a) and Canada (Rush 1981) are inconsistent. It appears that acute and severe undernutrition during pregnancy does impair fetal growth and that this effect disappears as soon as dietary intake improves, as was the case during the Dutch hunger winter (Stein & Susser 1975). Fetal growth

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retardation can be reduced by an increase in energy intake during pregnancy as shown in The Gambia (Prentice *et al.* 1983).

In most developing countries undernutrition is of a chronic and marginal nature. It is still not clear who would benefit most from costly supplementary feeding programmes in such conditions (Habicht & Yarbrough 1980; Rush 1983b). The question relates to screening of recipients for proper targeting of dietary interventions as well as to the desired outcome of the intervention, namely birthweight, peri- and neonatal mortality, breast-milk yield, postnatal growth, the mother's nutritional status or even her physical (economic) activity.

The importance of the subject was considered to justify a controlled trial in a community exposed to chronic undernutrition, which as far as possible takes account of the methodological weaknesses of the previous studies.

The present study, conducted in East Java, Indonesia, was designed to test the effect of energy supplementation in the last trimester of pregnancy on birthweight, breast-milk yield and postnatal growth. This article deals with birthweight.

Patients and methods

Study population

Three study villages were chosen from a district in a semi-arid zone, located along the southern coast of Madura known to be nutritionally vulnerable (Kardjati *et al.* 1983). The main occupations are agriculture, fishery and salt production.

Experimental design

The study design was a randomized, controlled trial. In group discussions the community was made aware of the importance of good nutritional health during pregnancy for the baby's size, health and survival. Households with pregnant women and traditional birth attendants were informed that the project offered free antenatal care, an energy-enriched 'jamu' (see *Supplements*) and curative care for pregnant women and their infants. In each village pregnant women who were willing to participate in the study were allocated to groups offered high or low energy supplements from 26 to 28 weeks gestation (assessed clinically). Each house in the village has been given a number during the cen-

sus at the start of the project. The household numbers were the basis for allocation using random number tables. While the study was not blind, the randomization was, since the characteristics of the pregnant women cannot be inferred from the household numbers. Sample size was calculated to detect a difference in mean birthweight between high and low energy-supplemented groups of 100 g at a significance level of 5% with a power of 80%, using a two-sided test ($n = 150$). It was assumed that the standard deviation of birthweight in this population was 300 g. To allow for drop-outs at least 250 women were enrolled in each group.

The experimental period (births between September 1982 and December 1984) was preceded by a baseline period (births between August 1981 and August 1982). The latter was meant to provide information of habitual dietary intake and birthweight distribution in the study population.

Supplements

From previous nutrition surveys in the same area it was known that habitual diets of pregnant women were mainly deficient in energy (Kardjati *et al.* 1983). It was therefore decided to provide a supplement which would permit an increase of their energy intake to the estimated energy requirement level. In practice this involved giving a supplement mainly as fat. To improve acceptance and to reduce sharing, a supplement was developed which could be taken with 'jamu'. This is a traditional, bitter-tasting beverage, an extract of herbs, commonly taken by women during pregnancy to maintain health and to ensure an easy delivery. A small amount of casein was added to the fat for technological reasons (i.e. the fat should be readily soluble in hot water like a coffee creamer) and to keep the protein-energy percentage of the home diet plus the supplement at 10% on average. The composition of the high and low energy supplement is shown in Table 1. Female fieldworkers, recruited and trained locally, delivered one package of the supplement each day at home. They waited till the women had consumed the amount they wanted, returning the leftovers to the field station to be weighed.

Table 1. Composition of the supplemental beverages

Ingredients		Daily serving	
High-energy	Low-energy	High-energy	Low-energy
50% fat*	50% casein	1.95 MJ	218 kJ (52 kcal)
10% casein	28% glucose	(465 kcal)	6.2 g protein in
40% glucose	22% Textaid	7.1 g protein in 75 g dry mix†	15 g dry mix†

* Sunflower oil: palm oil = 1:1.

† To be dissolved in 200 ml of boiling water

Anthropometry

The schedule of data collection is presented in Table 2. At monthly intervals eligible couples were visited at home for case-finding of pregnant women. On these occasions all women were weighed with a bathroom scale to obtain pre-pregnant weight. When found pregnant, women were referred to the antenatal clinic in each village for obstetric interview and examination and to be weighed and measured. Maternal anthropometric measurements were taken in duplicate at each monthly antenatal visit, using standardized techniques (Weiner & Lourie 1969). Quality control as described by Habicht (1973) and checking of equipment were done at 3-monthly intervals. Birthweight was measured within 24 h with a Salter scale of 10 kg to the nearest 50 g.

Table 2. Schedule of activities and data collection

Type	Period	Frequency	Method
Enrolment of pregnant women	B + E	Monthly	Home visit
Obstetric history and examination	B* + E	Monthly	Interview and examination at project's clinic
Anthropometry			
Prepregnant weight	B* + E	Monthly	Home visit
Height (1×), weight, upper arm circumference, four skinfolds	B* + E	Monthly	Clinic
Home diet	B + E	4×	Three-day food weighing
Distribution of supplement	E	Daily	Home visit
Birthweight	B + E	Within 24 h	Home visit
Socio-economic variables	E	1×	Clinic or home visit
Agricultural production	B + E	Monthly	Investigators' records

B, Baseline period; E, experimental period.

* Incomplete coverage.

Food consumption

For each woman 3-day food weighings were scheduled at pregnancy month 5–6 and then monthly. The female fieldworkers stayed in the household at cooking and eating times for 3 consecutive days to weigh all foods cooked and eaten by the woman. Snacks eaten between meals were recorded at the next mealtime. The amounts of raw food consumed per day were converted into energy and nutrients, using the Indonesian and the FAO Food Composition Table for use in East Asia (FAO 1972).

Socio-economic status of households

A socio-economic score for households was based on a summation of weighted scores for education of husband and wife (maximum 10 points), type of housing (maximum 20 points), type of furniture (maximum 5 points), ownership of livestock (maximum 10 points), ownership of a motorcycle, radio, jewellery, etc. (maximum 30 points).

Data analysis

Pregnant women took widely differing amounts of the supplements offered to them at home. Apart from testing outcome per experimental group, account had to be taken of the degree of compliance. Three subcategories per experimental group were used, namely women who consumed less than 45 packages, 45–89 packages and 90 or more packages. In the high energy

group the extra energy consumed corresponds to <87.6 MJ (20 925 kcal) (HE-1), 87.6–175.2 MJ (20 925–41 850 kcal) (HE-2) and >175.2 MJ (41 850 kcal) (HE-3) respectively. In the low energy group the corresponding figures are <9.8 MJ (2340 kcal) (LE-1), 9.8–19.6 MJ (2340–4680) (LE-2) and >19.6 MJ (4680 kcal) (LE-3). For the analysis of the effect on birthweight, an analysis of covariance was made. Birthweight was the dependent variable. The factors of interest in this study were high and low energy supplementation, number of packages consumed, parity (primi- vs multiparae) and sex of the infant. Height was treated as covariable. To assess which factors were associated with birthweight for each experimental group, a stepwise multiple regression analysis was performed. In this analysis, variables were included or excluded one by one, depending on whether they gave a significant contribution in explaining the residual variation. A variable was included if its contribution was significant at the 10% level and excluded if its contribution was no longer significant at the level of $P = 20\%$.

The study was approved by the Directorate of Research, Ministry of Health, Indonesia, and the peer review group for medical research of the Royal Tropical Institute. Both have dealt with the ethical considerations.

Results

Baseline period

(1) *Home diet.* The staple foods of the Madurese are rice, maize and cassava, eaten as a mixture of

rice–maize or maize–cassava, depending on the season. Fish is eaten daily in small amounts. The menu is complemented by gourd-like vegetables. Leafy vegetables and pulses are only available during a few months of the year.

The mean energy and nutrient intakes of pregnant women in the baseline period are shown in Table 3. Habitual diets were particularly low in energy and marginal in protein. Intakes were similar between 6 and 9 months gestation and even lower in early pregnancy (Kardjati 1985). Energy intake was equal to or above the recommended allowances, formulated for Indonesian women (LIPI 1983) in about 10% of all 3-day food weighings.

If a level of 159 kJ (38 kcal) per kg bodyweight per day is taken as the requirement (Calloway *et al.* 1980; Whitehead *et al.* 1984), namely 7.2 MJ (1710 kcal) for a woman with a prepregnant weight of 45 kg, about 20% of the 3-day food weighings reached this level. On average, protein intake was slightly below the requirement level for Indonesian women (40 g) but about 20% of the intakes were above the recommended allowance of 50 g (Fig. 1). However, protein contributed 10% of the energy, and protein utilization improves if energy requirements are met.

(2) *Birthweight distribution.* In the baseline period 481 women gave birth to 479 singletons of which 11 were stillborn. Of the two sets of twins, one pair was stillborn. Birthweight was recorded in 419 liveborn singletons (87%). In the cultural setting it was not possible to weigh stillborn babies. The mean birthweight of liveborn singleton infants was 2835 g when two births of

Table 3. Daily energy and nutrient intake of 360 pregnant women in the baseline period

Energy/nutrients	Mean	(SD)	Median	RDA*
Energy				
(MJ)	6.28	(2.1)	6.0	8.73
(kcal)	1500	(499)	1434	2085
Protein (g)	41	(13)	40	50
Fat (g)	18	(10)	16	—
Retinol eq. (µg)	443	(345)	377	650
Thiamin (mg)	0.83	(0.32)	0.76	1.0
Riboflavin (mg)	0.36	(0.12)	0.34	1.3
Iron (mg)	6.7	(2.3)	6.3	30.0
Calcium (mg)	167	(128)	137	600

Total number of 3-day weighing periods = 970.

* Recommended daily allowance for Indonesian pregnant women as formulated at the National Workshop on Food and Nutrition, LIPI, Jakarta, 1983.

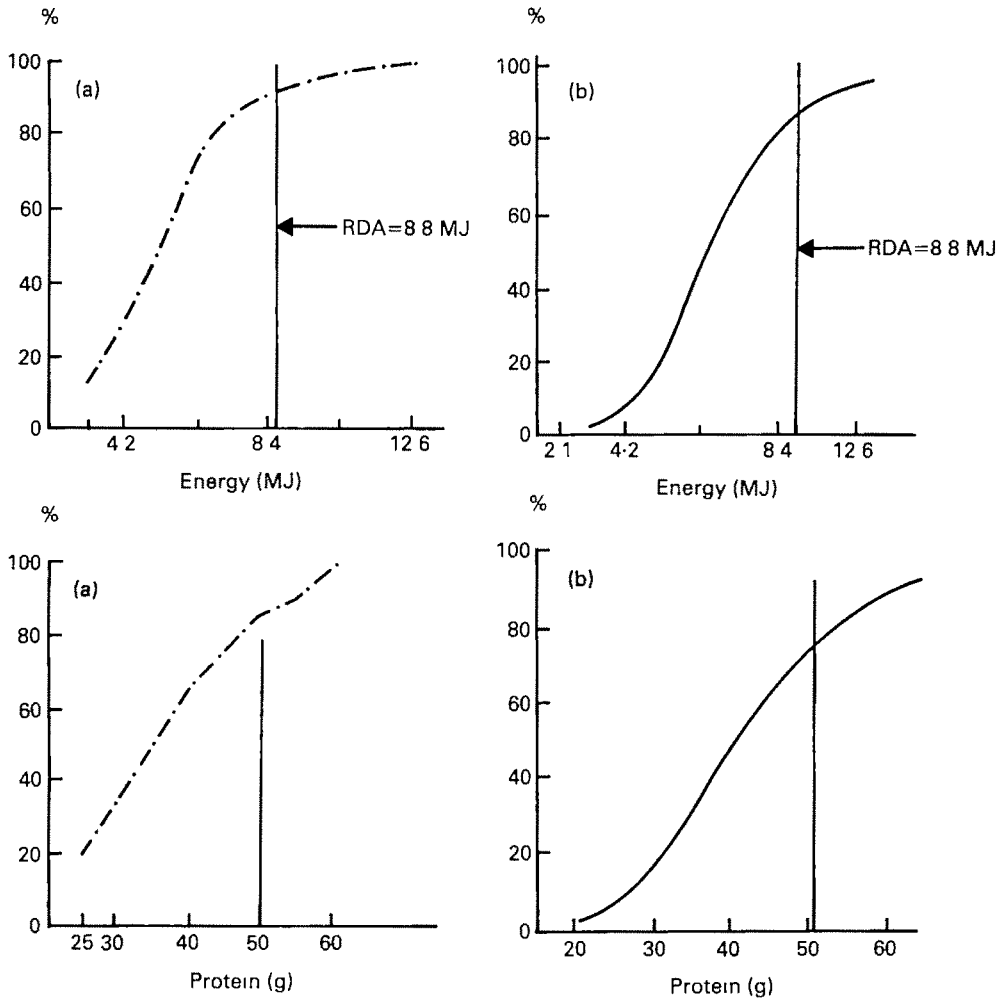


Fig. 1. Cumulative percentage frequency distribution of daily intake of energy and protein in the baseline (a) and experimental (b) periods. RDA, Recommended dietary allowance.

1000 g and 1500 g respectively were excluded (Table 4). For first-born babies mean weight was 2720 g and for babies of birth order 2 and higher 2850 g (not shown in Table). The incidence of LBW (less than 2500 g) was 12.2%. The birthweight distribution tended to be Gaussian.

Apparently mean birthweight was in agreement with the low dietary intake during pregnancy. A bivariate analysis of data from the sample with at least three food-weighing periods ($n = 190$) between 6 and 9 months gestation gave the following results:

$$\text{Birthweight} = 2607 + 0.239 \text{ kcal} - 3.48 \text{ protein, g}$$

$$R_2 = 0.035, F_{2:176} = 3.25, P = 0.040.$$

Energy and protein intake therefore influenced

birthweight significantly but explained only a small portion of the variation. From the analysis one can expect an increase in mean birthweight of 100 g per additional 500 kcal per day at equal level of protein intake.

Experimental period

(1) *Characteristics of pregnant women.* Women in Madura as a whole and in the study villages are housewives and productive labourers. As wives of farmers they assist in planting, weeding and harvesting. Fishermen's wives repair nets, go house-to-house or to the market to sell fish. Some women are petty traders, making and selling snacks. In households having cattle, women and children are usually responsible for collect-

ing fodder. Women continue with their regular work during pregnancy. Only in the last month before delivery might they reduce work which entails walking long distances.

Based on a time and motion study in a subsample of pregnant women, it was estimated that pregnant women spend about 18 h per 24 h sitting or sleeping, 3 h on food preparation, 1 h on domestic work and child-care and 1–2 h on hard physical work, such as agricultural work, selling fish, cutting grass and fetching water. Hence, the activity of these women can be considered light to moderate (Kardjati 1985).

The characteristics of pregnant women in the six subcategories HE-1–3 and LE-1–3, that is the high and low energy groups according to degree of compliance, and of non-compliers, are shown in Table 5. They appear to be comparable, except for the subcategory LE-1 with a higher pre-pregnant weight. Total weight gain during pregnancy was on average 7.1 kg in the HE group, 6.4 kg in the LE group and 6.3 kg in the non-compliers group (not shown in Table). The average socio-economic score (SES) for the two experimental groups was 30. Since the maximum possible SES is 75, households in the study population were generally poor.

(2) *Daily energy and protein intake from the home diet.* In a subsistence economy, fluctuations in food production and income are common as agriculture, salt production and to a lesser extent fishery are influenced by the rainy seasons. The good harvests and an abundant salt production from September 1982, coinciding with the start of the experimental period and continuing throughout 1983, were reflected in the energy and protein intake from home diets. The erratic rains in 1984 resulted in harvest failures and less opportunity for salt winning. These coincided with a drop in food consumption (Fig. 2). Consequently, the mean energy and protein intake from home diets was higher in the experimental period than in the baseline period, but they were similar for the HE, LE and non-complier groups. Thus no replacement effect was noted, nor was there an indication of self-selection by type of supplement. On the other hand, within experimental groups self-selection did occur. The subcategories, which consumed the highest amounts of supplement, namely HE-3 and LE-3, appeared to have the poorest home diet (Table 6).

(3) *Birthweight.* A total of 753 infants was born in the experimental period, among which there were six pairs of twins. Among the 717 liveborn singleton babies birthweight was measured in 687 (95%). As in the baseline period the birthweight distribution appeared to be Gaussian. Mean birthweight of the total sample (all experimental groups combined) was 100 g higher than in the baseline period, i.e. 2933 g. The corresponding figures for the HE, LE and non-complier groups were 2908 g, 2948 g and 2948 g respectively. The incidence of LBW was 9.5% (Table 7). Hence, energy supplementation had no apparent effect on mean birthweight. It seems, however, that supplementation was beneficial if it was targeted to the most in need. The subcategories HE-3 and LE-3 had the lowest intakes from home diets (Table 6). Group HE-3, which took the largest amount of supplementary energy also had the highest mean birthweight in both males and females (Table 8). In the LE-group one might expect a downward gradient in birthweight but this was limited to female babies.

Since the effects of different variables are interrelated, in the final analysis of the central hypothesis a stepwise, multiple regression analysis was performed per experimental group. The variables under consideration were: sex of the infant, parity (primi- vs multiparas), mother's height (cm), pre-pregnant weight (kg), mean energy intake from home diet, supplement (HE and LE), quantity of supplement consumed (number of packages: <45, 45–69, 70–79, 80–89, 90–99, ≥ 100).

In the LE group birthweight was only significantly influenced by maternal height as suggested by the following equation:

$$\text{Birthweight} = 101 + 18.9 \text{ height.}$$

$$r = 0.26, R_2 = 6.5\%.$$

In the HE group pre-pregnant weight ($r=0.26$) and number of packages consumed ($r=0.13$) influenced birthweight:

$$\text{Birthweight} = 1913 + 22.0 \text{ pre-pregnant weight} \\ + 27.1 \text{ quantity class.}$$

Discussion

Mean birthweight in the study population is within the range usually found in developing countries where undernutrition exists (Belizan *et al.* 1978; Villar & Belizan 1982). The percentage

Table 4. Birthweight distribution of singleton livebirths in the baseline period

Birthweight class (kg)	n (total)	Cumulative percentage frequency distribution		
		Males	Females	Total
<2.0	6	1	2	1.4
2.0–	45	11	13	12.2
2.5–	201	58	62	60.4
3.0–	144	94	95	95.0
3.5–4.1	21	100	100	100
All classes:				
n	417	218	199	
Mean (g)	2835	2838	2832	
SD (g)	363	360	367	

Two babies with birthweights of 1000 g and 1500 g respectively were excluded.

Table 5. Characteristics of pregnant women in the experimental period by subcategory (of singleton births)

Characteristics	High-energy group			Low-energy group			Non-compliers (n = 204)	Total (n = 741)
	1 (n = 55)	2 (n = 132)	3 (n = 85)	1 (n = 53)	2 (n = 105)	3 (n = 107)		
Age (years)								
n	54	131	85	52	105	107	190	724
Mean	24.4	25.9	24.5	25.7	25.9	24.9	26.5	25.6
SD	5.6	5.6	5.0	6.8	6.6	6.0	5.9	5.9
Birth order								
n	54	131	85	52	105	107	190	724
First (%)	28	15	20	25	24	17	21	20
2–7 (%)	63	77	79	65	72	76	69	73
8+ (%)	9	8	1	10	4	7	10	7
Birth interval (months) (since last delivery)								
n	30	92	50	29	58	71	131	461
Mean	28.4	28.2	30.7	27.3	29.9	29.6	29.4	29.2
SD	9.8	10.2	10.4	10.4	10.3	11.6	10.6	10.5
Height (cm)								
n	52	130	85	52	105	107	68	599
Mean	151.5	150.9	150.1	150.5	150.9	150.3	149.8	150.6
SD	5.2	4.9	4.7	5.6	5.0	5.2	4.8	5.0
Prepregnant weight (kg)								
n	38	100	64	36	76	84	141	539
Mean	42.2	41.8	42.2	44.5	42.8	42.4	43.2	42.6
SD	4.2	4.2	5.1	4.6	5.7	5.0	4.7	4.8
Prepregnant (W/H ²) (kg/m ² × 10)								
n	35	98	64	35	76	84	49	441
Mean	183	183	187	196	188	187	190	187
SD	18	17	22	19	22	19	19	20
Socio-economic score								
n	55	125	84	52	102	104	201	723
Mean	31.9	30.6	30.4	28.8	30.8	30.0	28.2	29.8
SD	10.4	8.3	7.7	6.8	8.6	6.6	9.1	8.4

Group 1: consumed less than 45 packages; group 2: consumed 45–89 packages, group 3: consumed 90 or more packages.

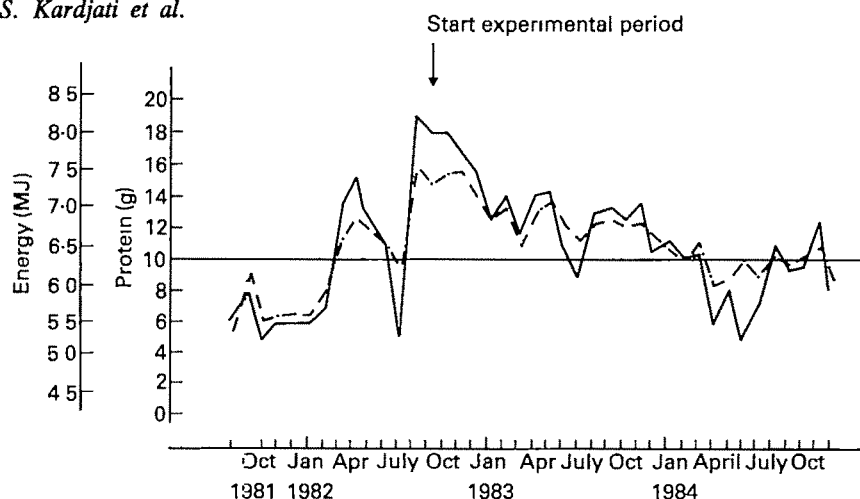


Fig. 2. Energy and protein intake from home diet by calendar month over the whole study period. —, Energy; ---, protein.

of LBW was relatively low but in agreement with figures from other areas in Indonesia (Bernard *et al.* 1980; Alisjahbana *et al.* 1983). In the experimental period the infant mortality rate was 121 per 1000 livebirths, first week and neonatal mortality rates were 28 and 52 per 1000 livebirths, respectively. The stillbirth rate was 33 per 1000 births (Kusin *et al.* 1988).

Many factors may influence fetal growth. Maternal dietary intake cannot be ruled out as a determinant of the outcome of pregnancy but it remains difficult to prove a direct and consistent relation between the two. The variation in results from previous dietary interventions can be attributed to differences in degree of undernutrition, methodological limitations and the manner in which confounding factors were taken into account (Stein *et al.* 1978; Habicht & Yarbrough 1980). In the East Java Pregnancy Study, advantage was taken of experience with other feeding trials.

Reliable data from the Nutrition Surveys (Kusin *et al.* 1979; Kardjati *et al.* 1983) and from the baseline period were available, which were indicative of chronic but moderate undernutrition during pregnancy. Habitual diets provided on average 6.3 MJ (1500 kcal) and 40 g of protein. No difference was found in home diets as pregnancy advanced (Kardjati 1985). Such a level of consumption was associated with an average prepregnant weight of 42 kg, i.e. a body mass index ($\text{weight/height}^2 \times 10$) of 187 at a mean height of 150 cm. While women appeared to be able to maintain an acceptable body mass index (WHO 1985) when not pregnant, a total

weight gain during pregnancy of 6–7 kg is obviously low. The bivariate analysis of energy and protein intake versus birthweight in the baseline period suggested that energy supplementation would be beneficial in terms of birthweight. The experiment followed a controlled, randomized design within villages, and the supplements were delivered at home to reduce self-selection. The choice of 'jamu' as a vehicle proved to be successful in avoiding replacement and leakage to other members of the family. Nevertheless, two confounding factors could not be controlled in free-living communities where participation was voluntary: the year-to-year fluctuations in home food supplies and the degree of compliance. Interpretation of the results should, therefore, be made against this background.

It is very likely that a cut-off point in daily energy intake exists, below which the risk of impaired fetal growth increases. In the Dutch hunger winter it corresponds with an intake of 1500 kcal per day (Stein *et al.* 1975). In the experimental period the majority of women consumed more than this amount (Figs 1 & 2). The absence of a difference in mean birthweight between the HE and LE groups as a whole may be attributed to a masking effect of the better home diet in the experimental period. A positive effect might be expected among women with the poorest home diets who took the greatest amount of supplementary energy. The observation that energy supplementation in the subcategory with the lowest home intake in the HE group was able to raise mean birthweight to the

Table 6. Daily intake in energy and protein from home diet of pregnant women in the experimental period by degree of compliance

Experimental subcategory	n	Energy intake		Protein intake (g)
		(MJ)	(kcal)	
High-energy				
1	163	7.19 (2.16)	1717 (516)	44.2 (12.9)
2	453	6.65 (1.91)	1590 (457)	41.6 (12.5)
3	321	6.62 (1.84)	1582 (440)	41.7 (11.7)
Low-energy				
1	163	6.76 (2.16)	1616 (515)	43.8 (13.9)
2	358	6.89 (1.87)	1645 (447)	43.4 (12.0)
3	392	6.46 (1.63)	1543 (388)	40.3 (10.4)
Non-compliers	369	6.45 (2.02)	1541 (483)	41.1 (13.8)

n, Number of 3-day food weighings.

1, Consumed less than 45 packages; 2, consumed 45–89 packages; 3, consumed 90 or more packages.

Results are mean (SD) values.

Table 7. Birthweight distribution of singleton livebirths in the experimental period

Birthweight class (kg)	High-energy		Low-energy		Non-compliers		Total	
	n	Cum %	n	Cum %	n	Cum %	n	Cum %
<2.0–	4	1.6	3	1.2	5	2.8	12	1.7
2.0–	25	11.2	21	9.5	7	6.8	53	9.5
2.5–	95	48.1	91	45.6	71	46.9	257	46.9
3.0–	118	93.8	109	88.9	77	90.4	304	91.1
3.5–4.1	16	100.0	28	100.0	17	100.0	61	100.0
All classes								
n	258		252		177		687	
Mean (g)	2908		2948		2948		2933	
SD (g)	397		392		440		407	

One baby with a birthweight of 1500 g was excluded.

level of the better-off in the same community supports this hypothesis, although Madurese women are smaller than the Dutch women. Similarly, one would expect a downward gradient in mean birthweight in the LE group, corresponding to a trend in home diets. This was indicated by the results but in female infants only.

Interestingly, in line with results from other feeding trials, the stepwise multiple regression analysis showed a modest effect of an increase in mean birthweight of about 50 g per (42 MJ) 10 000 kcal extra intake. The same analysis indicates the importance of prepregnant weight as a determinant of birthweight. It makes physiological sense that maternal energy reserves are used to buffer deficits in periods of need. The cut-off

point of 40 kg appeared to be the risk level of LBW in this community (Kardjati 1985).

To put the results in broader perspective, in the study population living in a semi-arid zone, a lean year such as in the baseline period would occur once in 4–5 years. Under 'normal' conditions the home diet would provide on average 6.4–7.2 MJ (1540–1720 kcal) and 41–44 g of protein per day from rice, maize, fish and vegetables. Such an energy intake would be less than the requirement for light to moderately active pregnant women formulated recently (WHO 1985). As mentioned earlier, women on average spent about 18 h sitting or sleeping, which may be reduced by about 2 h in the periods of peak activity in the agricultural cycle. Short-term energy supplementation during pregnancy in

Table 8. Mean and standard deviation of birthweight by subcategory of experimental group (birth order of the infants combined)

Sex	No of packages consumed	High-energy group			Low-energy group			Non-compliers			Total		
		<i>n</i>	Mean	(SD)	<i>n</i>	Mean	(SD)	<i>n</i>	Mean	(SD)	<i>n</i>	Mean	(SD)
Males	45	29	2935	(417)	26	2962	(496)						
	45-89	75	2890	(422)	54	2986	(371)	90	3011	(436)	368	2966	(406)
	90	39	2985	(316)	55	2980	(378)						
Females	45	22	2898	(357)	24	2961	(478)						
	45-89	51	2796	(423)	44	2958	(407)	87	2884	(436)	319	2896	(404)
	90	42	2954	(375)	49	2847	(305)						
Males + females	45	51	2919	(389)	50	2962	(482)						
	45-89	126	2852	(423)	98	2973	(386)	177	2948	(440)	687	2933	(407)
	90	81	2950	(346)	104	2917	(350)						

HE-1/LE-1. consumed less than 45 packages; HE-2/LE-2: consumed 45-89 packages, HE-3/LE-3. consumed 90 or more packages

these 'normal' years did not result in an improvement of birthweight. Several reasons are plausible. (1) The formulated requirements may be too high. Recent data from Cambridge suggest that optimal reproductive performance can be attained with a daily intake of about 8.37 MJ (2000 kcal), i.e. 840-1260 kJ (200-300 kcal) less than the supposed requirement for British women (Whitehead *et al.* 1984). (2) Maternal height may be a limiting factor (Thomson *et al.* 1968; Dougherty & Jones 1982). (3) The reduced placental blood flow due to working in a standing position under tropical temperatures places a constraint on optimal fetal growth (Briend 1980; Tafari *et al.* 1980). (4) In communities where women remain engaged in productive activities throughout pregnancy, the extra calories provided may be used for more physical work.

A striking difference between the INCAP study in Guatemala and the present one is the effect on the incidence of LBW (Habicht 1974; Lechtig *et al.* 1975a). It was very significant in the Guatemalan study, where the incidence before supplementation and in the general population ranged from 22 to 41%, but small and not significant in the East Java population with an incidence of 9-14%. This difference is likely to be due to the birthweight distribution. At a comparable mean birthweight, such a high incidence of LBW in Guatemala points either to a skewed distribution at the lower end or to the existence of a poor group within the 'normal' group (Lechtig *et al.* 1975b; Marks 1985).

The positive effect in the INCAP study was

mainly confined to the needy group, which appears to be in agreement with our findings.

In this paper we have not considered the possible effect of energy supplementation on maternal nutritional status.

With regard to birthweight, our results suggest that in communities exposed to chronic but moderate undernutrition without pockets of extreme poverty, energy supplementation for pregnant women may be most effective if targeted to lean seasons and/or to women with a low prepregnant weight.

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Ovarian size in postmenopausal women

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Summary. Ovarian volumes have been determined by pelvic ultrasonography in 2246 apparently healthy postmenopausal women of whom 2221 were included in the statistical analysis. Factors associated with gonadal size have been identified, and reference ranges for derived indices have been determined for use (in association with criteria for abnormal morphology) in a screening programme for ovarian carcinoma. The right ovary was present in 98.9% of subjects and the left in 99.1%. The mean (SD; range) of right and left ovarian volumes were 3.58 (1.40; 1.00-14.01) and 3.57 (1.37; 0.88-10.9) ml respectively. Significant predictors of ovarian volume were years since the menopause, weight, parity, age at menopause, a history of hormone replacement therapy, and previously diagnosed breast cancer. Abnormal ovarian volumes were assessed from a score equal to the (observed mean log volume (MLV) minus the predicted MLV)/0.327. A simplified nomogram has been prepared for routine clinical use. The relative abnormality of one ovary was assessed from a ratio score equal to $\log_e$ (larger ovarian volume/smaller ovarian volume)/0.211 compared with the 99th centile for the Gaussian distribution.

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In a preliminary communication we reported that real-time mechanical sector scanning can be used to determine ovarian morphology and volume in peri- and postmenopausal women (Campbell *et al.* 1982). All ovaries assessed to be of normal morphology by ultrasound were found to be so at subsequent laparotomy, and there was satisfactory agreement between the volumes as determined by sonar and by direct measurement at the time of surgery over the range 1-10 ml.

We were so encouraged by these results that a prospective study was designed to assess real-time mechanical sector scanning as a screening procedure for the detection of early ovarian neoplasia. The ultrasound technique has been published in detail and preliminary data clearly indicated that malignant neoplasia could be detected by changes in ovarian morphology or volume or both (Goswamy *et al.* 1983). More recently the approach has been used by other workers (Andolf *et al.* 1963).

The morphological abnormalities were described, but it became apparent that a large series of healthy women should be studied to establish reference ranges for ovarian volume. In this paper we present results from 2221 postmenopausal women who had undergone a natural menopause and were judged to have normal ovaries on examination with ultrasound. We have examined the factors which affect ovarian volume and devised methods for detecting factor-specific enlarged ovaries.

Patients and methods

Women were self selected in that they responded to advertisements placed within King's College Hospital or in the local or national press. A medical referral was not required. Any woman over 45 years could request a scan; women younger than 45 were also scanned if there was a personal or family history of certain types of cancer. We assigned women to one of two groups: natural postmenopausal (intact uterus, age ≥ 35 years, at least 1 year since last menstrual period), or artificial menopause (hysterectomy or radiation menopause). Data from the first group only are described here.

Women contacted the clinic and were given an appointment. They were instructed to drink two pints of fluid 1 h before ultrasound examination. By displacing the intestinal loops, the fluid-filled bladder provides a sonic window which enhances display of the pelvic organs (Donald 1963). Before examination the women completed a questionnaire designed to provide demographic data and a relevant personal and family medical history.

A morphologically normal ovary has a uniformly hypo-echoic structure like the uterus, an ovoid shape and a smooth echogenic outline (Goswamy *et al.* 1983). Transverse and longitudinal scans were performed to assess ovarian morphology. The maximum transverse (D_1), antero-posterior (D_2) and longitudinal (D_3) diameters of each ovary were measured and the ovarian volume was calculated using the formula

$$\text{Volume} = (\pi/6) \times D_1 \times D_2 \times D_3.$$

Statistical analysis

All categorical (e.g. social class) variables were tabulated, and means, standard deviations and ranges calculated for numeric variables (e.g.

weight). For analysis a no answer to a categorical variable was classified as 0 and a yes answer as 1. Ovarian volumes were transformed to natural logarithms before analysis because a preliminary study of the data revealed that they had an approximately log-normal distribution.

It was anticipated that ovarian volume might be affected by a number of anthropometric and other variables. Multiple linear regression analysis was applied to mean log ovarian volume (MLV) to identify the influential factors. Backwards stepwise regression was performed, i.e. all variables of interest were included at the initial stage, and were eliminated singly if there was no significant reduction in the variance of the log ovarian volume at the 1% level (F-test). The stepwise procedure was repeated until no more variables could be eliminated, and the final model confirmed as the best by all-subsets regression analysis using the computer program BMDP9R (Brown 1981).

Regression analysis was also performed on the log ratio of the larger ovarian volume to the smaller ovarian volume, since one relatively large ovary could also indicate abnormality even in the presence of a mean volume in the normal range.

All calculations, except all-subsets regression, used a microcomputer with the statistics package SPP (Royston 1985).

Definitions of some categorical and numeric variables

Abdominal surgery was defined as laparoscopy or laparotomy for any condition and includes laparoscopic sterilization, appendicectomy, cholecystectomy, etc. The family history of cancer was restricted to the maternal side and there was no differentiation between first, second and third degree relationships. A history of endocrinopathy included thyroid, adrenal and pituitary diseases. A history of infertility was defined as difficulty in conceiving for at least 1 year. A history of oral contraceptive use and hormone replacement therapy was defined as hormone ingestion for at least 1 year with medical advice being sought. Social class was defined according to the Office of Population Censuses and Surveys. A woman who was separated from her husband was classified as married.

Results

Data from 2246 women were available for analysis. Twenty-five subjects were excluded (16 because the age at menopause was not known or was <35 years, 4 because of difficulty in viewing the ovaries for clinical reasons, and 5 because the ovaries were so small that accurate measurements of volume could not be made). Of the remaining 2221 subjects the mean age was 56.3 (SD 5.2; range 44–78) years, the mean height was 1.625 (SD 0.066; range 1.245–1.854) m; and the mean weight was 63.7 (SD 9.7; range 38.0–120.9) kg.

Summaries of data on the categoric and numeric variables are shown in Tables 1 and 2 respectively.

Most women screened were married (86.2%), underwent menarche between 13 and 15 years of age (67.1%), and had a normal duration of menstrual blood flow from 3 to 8 days (90.2%). Overall, 11% had never been pregnant, 54.7% had two or three children and 5.6% had five or more children, 13% gave a history of infertility. The

classification of patients by socio-economic class was similar to that in the general population of women in the given age groups (Office of Population Censuses and Surveys).

Overall, 95% had no family history of cancer and 37.8% had just one relative who had suffered from a malignant disease. In keeping with the relatively recent release of oral contraception for general use, 77.0% of the women screened had never used this method and most users (98.0%) had been prescribed the combined oral contraceptive pill. Overall, 83% had never received hormone replacement therapy for menopausal symptoms; 155 women (7%) gave a history of unopposed oestrogen therapy, while 199 (9.0%) had been on combined/sequential oestrogen and progestogen therapy. Of the study population 3.2% (71 women) reported having had breast cancer.

Mean ovarian volume

Overall, the right ovary was observed in 98.9% and the left in 99.1% of the women. Mean (SD;

Table 1. Some categoric variables of 2221 postmenopausal women in the study

Variate	Yes <i>n</i> (%)	No <i>n</i> (%)	Unknown <i>n</i> (%)
Abdominal surgery	849 (38.2)	1372 (61.8)	0 (0.0)
Breast cancer	71 (3.2)	2150 (96.8)	0 (0.0)
Breast lumps (benign)	367 (16.5)	1854 (83.5)	0 (0.0)
Current smoking	433 (19.5)	1786 (80.4)	102 (0.1)
Diagnosed diabetes	15 (0.7)	2206 (99.3)	0 (0.0)
Dilatation and curettage	1033 (46.5)	1188 (53.5)	0 (0.0)
Family history of cancer	102 (4.7)	2080 (95.3)	0 (0.0)
Gynaecological cancer	13 (0.6)	2208 (99.4)	0 (0.0)
History of endocrinopathy	112 (5.0)	2109 (95.0)	0 (0.0)
History of infertility	297 (13.4)	1924 (86.6)	0 (0.0)

Table 2. Data for some numeric variables within the study population of 2221 postmenopausal women

Variate	<i>n</i>	Mean	SD	Range
Age (years)	2221	56.3	5.2	44–79
Age at menopause (years)	2221	49.6	4.0	35–62
Duration of HRT (years)	369	2.4	2.2	<1–14
Duration of OC (years)	510	5.3	4.7	<1–20
Gravida	2219	2.5	1.7	0–14
Height (m)	2220	1.625	0.066	1.245–1.854
Parity	2219	2.1	1.4	0–9
Weight (kg)	2220	63.7	9.7	38–121
Years of infertility	296	5.8	3.8	1–29
Years since last pregnancy	1949	24.1	6.9	5–47
Years since menopause	2221	6.7	5.3	1–37

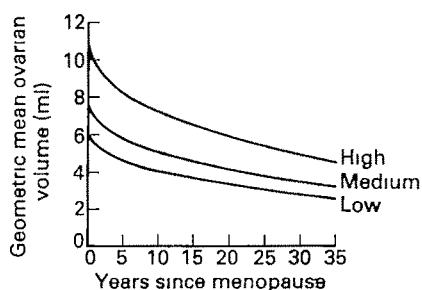


Fig. 1. The 95th centiles for high, medium and low expected ovarian volumes (geometric means) based on data from 2221 postmenopausal women.

range) for right and left ovarian volumes were 3.58 (1.40; 1.00–14.01) and 3.57 (1.37; 0.88–10.90) ml respectively. As indicated above, the distribution of ovarian volumes was skew and the mean log volume (MLV) and log ratio (LR) were calculated for regression analysis:

$$\begin{aligned}\text{MLV} &= 0.5 \log_e (\text{right volume} + \text{left volume}) \\ \text{LR} &= \log_e (\text{right volume}/\text{left volume}).\end{aligned}$$

If only one ovary was present, its log volume was used as an estimate of MLV but the LR could not be calculated.

The results of regression analysis on MLV are given in Table 3. Predictors above the dashed line form the final model, and their regression coefficients and standard errors are presented. Those below the line were not statistically significant, and the direction of their weak association with MLV is given as plus if they tended to increase volume and minus otherwise. The *F* ratios are 'F-to-remove', i.e. measure the significance of each predictor in the model in terms of reduction of the variance of MLV explained if the terms were removed one at a time. The final model had an index of determination (R^2) of 0.178. The residual SD, representing scatter about the regression line, was 0.327 log ml.

Significant predictors of ovarian volume were a transformation (LYRS) of years since the menopause (see Table 3), the number of livebirths (0, 1, 2 or more), the age at menopause, and a history of hormone replacement therapy and/or breast cancer. To calculate a score for abnormal volume a predicted MLV is calculated according to the regression coefficients in Table 3, i.e.

$$Y = B_0 + B_1X_1 + B_2X_2 + B_3X_3 + B_4X_4 + B_5X_5 + B_6X_6$$

where Y = predicted MLV,

B_0 = intercept,

$B_1, \dots, B_6$ are the regression coefficients (Table 3),

$X_1, \dots, X_6$ are the regression variables (Table 3), and used in the following expression:

$$\text{score} = (\text{observed MLV} - \text{predicted MLV}) / 0.327$$

where 0.327 is the residual SD.

The score should approximate a Gaussian distribution with mean 0 and SD 1. The observed MLV can be considered large if the score exceeds 1.645 (i.e. the 95th centile), and almost certainly large if it exceeds 2.58 (i.e. the 99.5th centile).

To simplify the application of the regression equation for use in clinics, in Fig. 1 we have plotted predicted 95th centiles of geometric mean volume (anti-log of MLV, equal to the square root of the product of right and left volumes) versus years since the menopause for three hypothetical situations, corresponding to high, medium and low expected volumes. The predictors are given in Table 4 where the values for age at menopause and log weight are the 95th, 50th, 5th, and 5th, 50th, and 95th centiles respectively. The percentages of patients whose ovarian volumes lay above the three lines in Fig. 1 were respectively 0.4, 4.9 and 18.1; thus the lines correspond to approximately the 99.5th, 95th and 80th centiles of our sample. It is suggested that a patient be classified as possibly or potentially abnormal if the geometric mean volume of both ovaries lies above the high line. If it is between the high and low line a volume score should be calculated as follows:

Example

The geometric mean ovarian volume of a 50-year-old woman (10 years since the menopause) weighing 70 kg was 8 ml. The calculated MLV was 2.0794. The subject had one child, a history of HRT, and no evidence or history of breast cancer. Accordingly:

$$\begin{aligned}\text{Predicted MLV} &= \\ &= 0.5824 (\text{intercept}) + -0.1197 \times 3.40 \text{ LYRS } [10/10 + \\ &\quad \log_e (1 + 10)] \\ &\quad + 0.3017 \times 4.25 (\log_e \text{ weight } 70) \\ &\quad + 0.0557 \times 1 (\text{parity } 1) \\ &\quad + -0.0088 \times 40 (\text{age at menopause}) \\ &\quad + 0.0828 \times 1 (\text{a history of HRT}) \\ &\quad + 0.1124 \times 0 (\text{no history of breast cancer}) \\ &= 1.2441 \\ \text{and the score} &= (2.0794 - 1.2441) / 0.327 \\ &= 2.5544.\end{aligned}$$

Table 3. Results of multiple regression analysis of mean log volume (MLV) Possible predictors below broken line had coefficients that were not significant at the 1% level. Final regression model ($n = 2221$ women) is given by first seven predictors.

Predictor	Regression coefficient	95% confidence interval	P
LYRS*	-0.1197	-0.132 , -0.107	<0.0001
Log _e weight (kg)	0.3017	0.21 , 0.39	<0.0001
Parity (0, 1, 2 or more)	0.0557	0.037 , 0.075	<0.0001
Age at menopause (years)	-0.0088	-0.0125, -0.0051	<0.0001
History of HRT (0=No, 1=Yes)	0.0828	0.046 , 0.119	0.0001
Intercept	0.5824	0.15 , 1.01	0.0001
Breast cancer (0=No, 1=Yes)	0.1124	0.035 , 0.190	0.0018
Abdominal surgery	+	Gynaecological cancer	+
Age at menarche	+	Height	+
Breast lumps (benign)	+	History of endocrinopathy	-
Current smoking	-	History of infertility	+
Dilatation and curettage	+	History of OC	-
Diagnosed diabetes	-	Marital status	+
Duration of HRT	+	Socio-economic group	+
Duration of OC	-	Years since first pregnancy	+
Family history of cancer	+	Years since last pregnancy	-
Gravida (0, 1, 2+)	+	Years of infertility	-

*LYRS = $Y/10 + \log_e(1 + Y)$, where Y is years since menopause.

HRT, Hormone replacement therapy; OC, oral contraceptive.

Table 4. Value of predictors of 95th centile of mean log volume (MLV) for high, medium and low expected volume. LYRS (see Table 3) is assumed to vary

Predictor	High	Medium	Low
Pred† + 1.645 × SD*	2.388	2.040	1.815
Log _e weight	4.413	4.143	3.921
Parity	≥2	≥2	0
Age at menopause	42.128	50.222	55.533
History of HRT	yes (1)	no (0)	no (0)
Breast cancer	yes (1)	no (0)	no (0)

*SD is the residual standard deviation from multiple linear regression.

†Pred is the predictor of MLV excluding LYRS.

HRT, Hormone replacement therapy

This value is above the upper limit of a 99% reference range for MLV and therefore may be indicative of ovarian enlargement.

Log ratio

Regression analysis of LR (log ratio of the larger to the smaller ovarian volume) showed that this index was independent of all the predictors tried. Its mean (n ; SD; range) was 0.002 (2178; 0.211; -0.983-1.289), and was not significantly dif-

ferent from zero using Student's one-sample t -test. The values followed an approximately Gaussian distribution. Thus a simple calculation is required to establish relative abnormality of one ovary compared with the other:

ratio score = $\log_e(\text{larger ovarian volume/smaller ovarian volume})/0.211$ where 0.211 is the residual SD.

The ratio score can be defined as abnormal if larger than an appropriate centile of the standard Gaussian distribution, e.g. 2.58 for the 99th centile; the score test is now two tailed.

Example

The right and left ovarian volumes were 12 and 7 ml respectively. Thus the ratio score is $\log_e(12/7)/0.211 = 2.554$. This value corresponds to the 99.5th centile of a Gaussian distribution and may therefore indicate some degree of abnormality.

Discussion

This is the first report of methods to detect abnormal size in one or both ovaries of postmenopausal women by real-time pelvic ultrasonography. Previous workers have measured surface areas (Paradisi *et al.* 1984) and ovarian volumes (Orsini *et al.* 1985) in menopausal women with regular ovarian cycles, and in patients with polycystic ovarian (PCO) disease. The mean ovarian volume for 25 premenopausal women was 7.94 (SD 2.3) ml and the range 5–15 ml. The corresponding values for 44 women with PCO disease were: mean 14.04 (SD 7.36; range 3–42) ml. Our values for 2246 postmenopausal women were: right ovary (mean 3.58; SD 1.40; range 1.00–14.01) ml and left ovary (mean 3.57; SD 1.37; range 0.88–10.90) ml, which do not differ significantly from the dimensions reported for 1016 women in the preliminary communication (Goswamy *et al.* 1983).

An analysis of the characteristics of the self-selected sample of women presenting for the screening test for early ovarian cancer indicated that it was similar to the general population of comparable age. This conclusion was reached after comparing the distribution of social class (and other variates such as marital status, parity and history of infertility) in the screened group with the corresponding figures for the general population (Office of Population Censuses and Surveys).

The results to date have shown that ovarian morphology and volume can be assessed in 99.8% of postmenopausal women who participated in the screening programme. This proportion is an improvement on our published results of 84.0% (Campbell *et al.* 1982) and 99.4% (Goswamy *et al.* 1983). We attribute this new success rate to our accumulated experience of the equipment and screening procedure.

An important initial finding was that the values of ovarian volume followed a log-normal distribution. Consequently, all subsequent analyses were undertaken on $\log_e$ data. The results of regression analysis indicated that the most impor-

tant predictors of ovarian volume were a transformation of the number of years since the menopause (i.e. $LYRS = Y/10 + \log_e(1 + Y)$, where Y is years since the menopause), the woman's weight, parity, age at menopause and a history of and/or breast cancer. The sharp decrease in ovarian size after the end of reproductive life is illustrated in Fig. 1, and it was observed that an early menopause was associated with smaller ovaries in subsequent life. The effects of body weight, parity and HRT were to increase ovarian size, and the results showed that once weight was known, height added very little information (Table 3). Women with just one pregnancy attaining viability had ovaries which may be enlarged by 6%. Multiparous women had ovaries that were approximately 12% larger. There was no additional effect of parity if the woman had more than two children compared with two. A surprising finding was the fact that 50% of all women who had received HRT to alleviate menopausal symptoms had been prescribed oestrogen therapy alone, although the beneficial and protective effects of progestogens on the endometrium have been widely promulgated (Whitehead *et al.* 1981). It is also of interest that a history of breast cancer was associated with an increase in mean ovarian volume of approximately 11%. Factors that did not exert a statistically significant effect on ovarian size include a history of contraceptive usage, a history of infertility, the presence of benign lumps in the breast, and a family history of cancer.

As a result of this study we conclude that real-time pelvic ultrasonography may be used to determine ovarian size and morphology in 99.8% of postmenopausal women. In addition, we recommend the use of a diagram (Fig. 1) to determine abnormal mean ovarian size in routine clinical practice. If the value for the geometric mean volume falls between the high and low lines then the score should be calculated and compared with an upper limit for the normal distribution. Similarly, the relative abnormality of one ovary compared with the other in the same woman can be assessed from the ratio score, which is independent of all the predictors of ovarian volume. These indices for abnormal ovarian size (geometric mean volume, mean ovarian score, ratio score) used in conjunction with criteria for abnormal morphology (Goswamy *et al.* 1983) are being used in a screening programme for early ovarian carcinoma (S. Campbell *et al.*, unpublished observations).

Although there is no evidence, to date, for differences in ovarian size between ethnic groups, the results from recent studies on variations in the size of human testes (Diamond 1986) suggest that each ovarian screening centre should establish its own reference ranges.

Acknowledgments

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The ultrastructural changes of the mucosa of blocked fallopian tubes

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Summary. The ultrastructure of the epithelium was studied in four blocked fallopian tubes which differed in the site of blockage and the degree of luminal distension. In all four specimens, there were large areas of the luminal surface which became barren and devoid of both the secretory and ciliated cells. Necrotic secretory cells and partially sloughed ciliated cells were observed in the remaining epithelium. Isolated patches of attenuated epithelium decorated with ciliated cells were sometimes found in the crypt of the mucosal folds. Results of this study suggest that pathological changes in the epithelium are common to tubes of both the terminal and cornual blockage.

Secretory and ciliated cells are the two primary cell types which make up the epithelium of the human fallopian tube (Clyman 1966; Critoph & Dennis 1977; Jansen 1980; Patek 1974; Patek *et al.* 1972a,b; Peters 1986). During the menstrual cycle, there are regular changes in the extent of ciliation, the proliferative activity and the secretory characteristics of the epithelium (Donnez *et al.* 1985; Verhage *et al.* 1979). Specific changes in the pattern of ciliation and the appearance of the secretory cells are also seen during the development of the tube and in the puerperium (Patek & Nilsson 1973; Seki *et al.* 1978). It is often emphasized that fertility is closely related to the pattern of tubal ciliation. However, it has been noted that in a study of simple hydrosalpinx, the pattern of ciliation was essentially normal (Patek & Nilsson 1977) and in

another study, the extent of deciliation of the hydrosalpingeal tube was not correlated with the postsurgical fertility of the patient (Fedele *et al.* 1984). The immobilization of ciliary action in tubes from patients suffering from Kartegener's syndrome is said to be associated with impaired fertility (McComb *et al.* 1986; Afzelius *et al.* 1978). In the present study, we have examined specimens from four blocked tubes with the site of blockage either at the terminal end or at the cornual segment. Special attention was paid to the ultrastructural changes in the epithelium which might be common to all these cases.

Patients and methods

Four women with a history of primary infertility of 4-13 years were included in this study. Two women had a salpingostomy for terminal blockage of the tube associated with hydrosalpinx. Tubal re-anastomosis was performed in the other two to bypass the cornual blockage. Biopsies of the fallopian tube were obtained from the site of obstruction before re-anastomosis by cutting with micro-scissors. For comparison a normal tubal specimen was obtained during tubal sterilization. The specimens were fixed in half-strength Karnovsky fixative for 12-24 h at 4 °C. The fixed specimens were then trimmed to

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expose the luminal surface of the tube. Further preparation of the specimen for transmission (TEM) and scanning electron microscopy (SEM) followed standard procedure. Briefly, the specimens were post-fixed in 1% osmium tetroxide and then embedded in Spurr plastics



Fig. 1. The epithelium of a normal fallopian tube viewed under the SEM showing the secretory cells which are covered with numerous microvilli and the ciliated cells. Bar = 10 μ m.

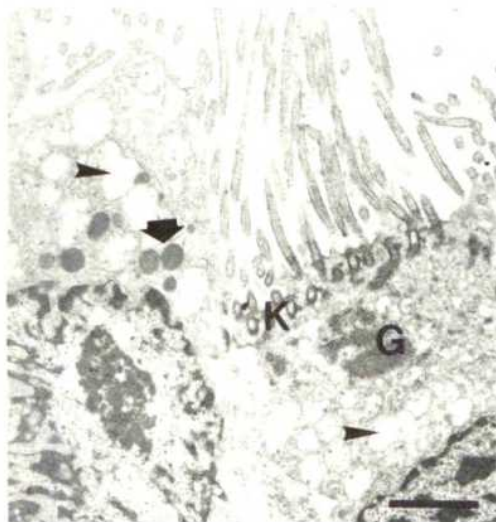


Fig. 2. Apical regions of a secretory cell and a ciliated cell in the normal fallopian tube epithelium. Dilated cisternae coated with amorphous materials are seen in both cell types (arrowheads). Secretory granules are found only in secretory cells (arrow). Glycogen granules (G) and kinetosomes (K) of the cilia are found in the ciliated cell. Bar = 1 μ m.

for sectioning, or critical-point-dried using Freon as the exchange fluid for SEM. Ultrathin sections stained with uranyl acetate and lead citrate were examined with a JEOL JEM-100CXII electron microscope operating at 60 kV, and the gold-palladium-coated specimens were examined with a JEOL 35-CF microscope operating at 15 kV.

Results

SEM examination of the luminal surface of the ampullary region of a normal tube revealed the presence of secretory and ciliated cells in the epithelium. The secretory cell typically had a bulbous apical surface which was covered with numerous microvilli (Fig. 1). Many secretory granules with dense inclusion and numerous smooth vesicles and dilated cisternae were found in the supra-nuclear cytoplasm of the secretory cells (Fig. 2). The irregular outline of the nucleus of the secretory cell was characteristic of those at the luteal phase of the menstrual cycle. The proportion of ciliated cells in the epithelium was estimated to be about 28% ($\pm 3\%$, scored in eight randomly selected areas in the ampullary region). The ciliated cells were found either as strands on the epithelium or more often as isolated cells interspersed with the secretory cells (Fig. 1). The dense tuft of cilia was connected basally to the kinetosome in the subapical cytoplasm (Fig. 2). Coated vesicles and dilated cisternae similar to those found in the secretory cells were also seen in the cytoplasm of the ciliated cells (Fig. 2).

Specimens from each type of blockage, two terminal blockages with hydrosalpinx and two cornual blockages with no luminal distension, were examined. In all four specimens, the ultrastructural appearance of tubal epithelium was similar. A common feature on SEM was the extensive effacement of the normal tubal epithelium, and the replacement by a cellular lining lacking the characteristics of either the secretory or ciliated cells (Fig. 3). This feature corresponded to the TEM appearance of a low cuboidal cell lining, which often showed a loss of nuclear polarity (Fig. 4). The luminal surface was covered by a thick mat of dense amorphous material (Fig. 4). Isolated residual patches of apparently normal tubal epithelium were seen, usually found in the crypt-like spaces formed by the mucosal folds (Fig. 3). In localized areas, the epithelium was replaced by a much flattened cell

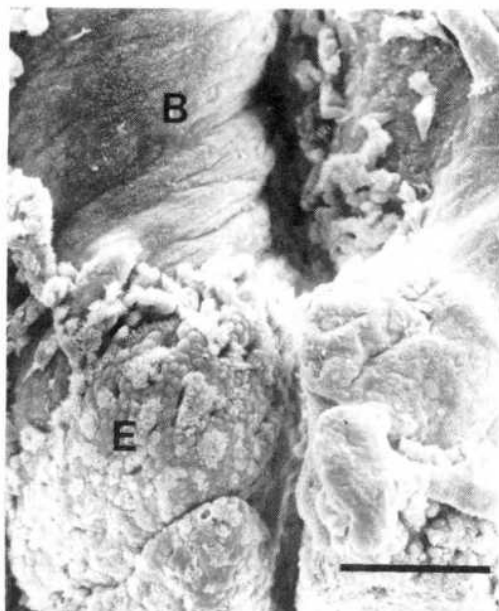


Fig. 3. The luminal surface of a fallopian tube with terminal blockage and hydrosalpinx showing the coexistence of a barren surface (B) and a small patch of normal epithelium (E). Some detaching ciliated cells are seen in the crypt of the mucosal fold (arrow). Bar = 100 μ m.

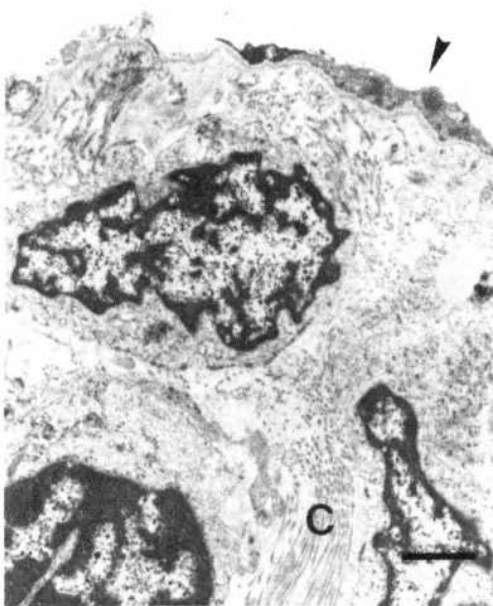


Fig. 4. The presence of a thick deposit of extracellular material (arrowhead) on the surface of the remaining epithelium of the barren surface and the abundant intercellular collagenous matrix between the epithelial cells (C). Bar = 2 μ m.

layer exhibiting sparse stunt microvilli (Fig. 5). In these deciliated areas, secretory granules were few and both the endoplasmic reticulum and golgi complex were poorly developed. Marked contrast was observed in regions demonstrating predominance of ciliated cells. In these ciliated areas, cells exhibited relatively well-preserved microvilli, although some degree of morphological changes was also observed. The secretory cells in these ciliated areas showed the formation of prominent cytoplasmic blebs on the apical surface (Fig. 6). The deciliated and ciliated areas were not always clearly demarcated. An admixture of intimately associated deciliated and densely ciliated epithelium was also observed (Fig. 7). Sloughing of lining cells, more often the ciliated type, was recognized in two specimens, one from each of the two types of blockage. The detaching cells had lost their intercellular junctions and cohesion with other cells and protruded into the tubal lumen (Fig. 7). It is unlikely that such sloughing of cells was due to trauma resulting from the dissection of specimens after fixation because the normal tubal specimen treated in a similar fashion did not show any abnormal changes in the epithelium. While many of these cells exhibited various stages of degeneration and necrosis, others demonstrated a well-preserved cytoplasmic structure (Fig. 8). The overall proportion of ciliated cells in the epithelium was 13% and 25% for the two specimens with hydrosalpinx, while this proportion was 6% and 2% for the two specimens with cornual blockage but no luminal distension.

Discussion

The present study, although based on only four examples, has clearly shown that tubal blockage was invariably associated with aberrant changes in the morphology of the tubal epithelium. Irrespective of the nature of the blockage and the degree of luminal distension, the ultrastructural appearance of the tubal epithelium was essentially similar for the four specimens. The most significant findings were that the normal tubal epithelium was lost over large areas of the tubal surface and was replaced by a non-secretory, flattened epithelium which was sometimes covered with amorphous material, and that ultrastructural appearance of the tubal surface was essentially similar among the different specimens with the exception of the proportion of

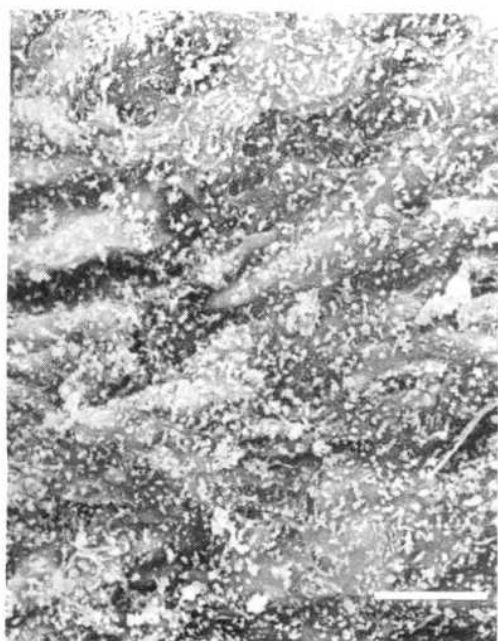


Fig. 5. The complete absence of ciliated cell in the epithelium of a tube blocked at the cornual segment with no tubal distension. Only flattened epithelial cells with sparse microvilli are found. Bar = 10 μ m.

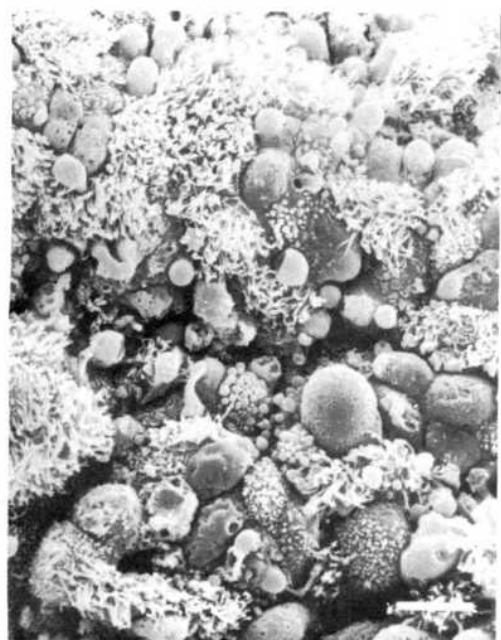


Fig. 6. The epithelial surface of another specimen from the same tube as in Fig. 5, showing a large number of ciliated cells and the blebbed secretory cells. Bar = 10 μ m.

ciliated cells in the epithelium. With the ciliated population, various intermediate stages leading to the total loss of the epithelium was seen in the specimens and the sloughing of ciliated cells was clearly demonstrated in the present study; whereas with the secretory cells, the absence of secretory granules and the prominent blebbing of the cells further revealed that the secretory activity of the tubal epithelium was also affected in these blocked tubes. In the present study, the normal tube used for comparison came from the tubal ampulla whilst the pathological specimens came from the ampulla in two and from the isthmic/interstitial segment in the other two. The overall similarity in the pathological appearance of the four specimens suggests that the results were not significantly affected by the site of origin of the biopsies although, strictly speaking, one should not compare tubal morphology of different segments since there is a well-documented increase in ciliation from ampulla to fimbriae in the fallopian tube (Patek *et al.* 1972a,b). Furthermore, the time during the menstrual cycle is another important determinant of tubal histology: secretory epithelium is best developed at mid-luteal phase and ciliary activity is highest at ovulation (Donnez *et al.* 1985; Verhage *et al.* 1979). The present study agrees with that of Patek & Nilsson (1977) in that a slight but not significant decrease in the ciliated population was found in hydrosalpingeal tubes: 13–25% compared with 22% in normal tubes. Inspection of the crypts of the mucosal folds revealed isolated patches of normal epithelium. This would suggest that the pathological or destructive process begins in the central lumen of the tube and spreads outwards. It is not possible to discern by ultrastructural examination of the site of obstruction whether changes in the tubal epithelium are the result of obstruction or of an inflammatory process. An examination of the epithelial changes in congenital obstruction of the fallopian tube may help to resolve these possibilities, but congenital obstruction is rare in women and perhaps it needs animal experimentation to generate such congenital blockage. Anatomical restoration of tubal patency often fails to achieve fertility and there is a high frequency of subsequent ectopic tubal gestation. Whether an entirely normal tubal epithelium would be restored after surgical anastomoses or salpingostomy is not known at present but if the aberrant epithelial morphology persists then the prognosis for postsurgical fertility would be low.

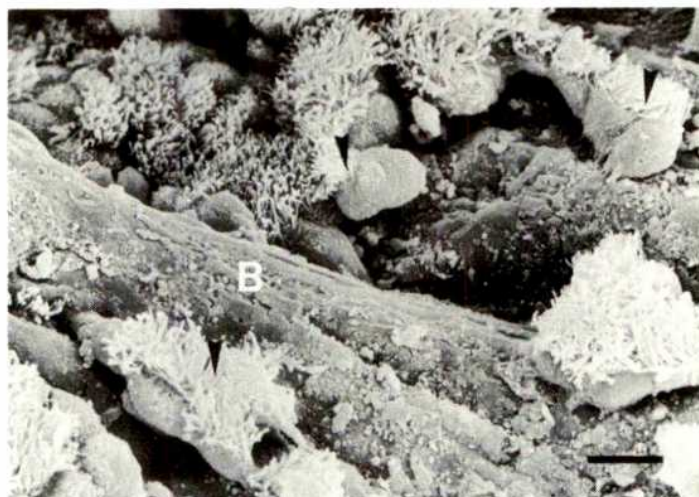


Fig. 7. A specimen obtained from a terminally blocked and distended tube showing the sloughing of ciliated cells (arrowheads) from the epithelium and the appearance of barren areas (B) of mucosal folds. Bar = 10 μ m.

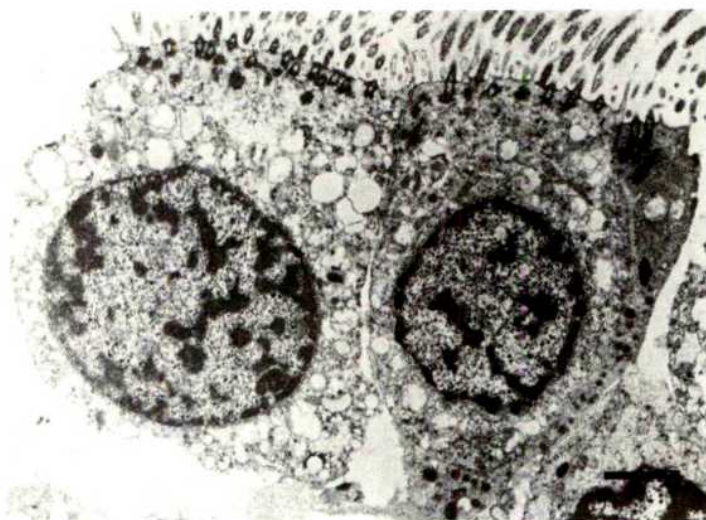


Fig. 8. Two detaching ciliated cells with healthy-looking cytoplasm. Many smooth vesicles and kinetosomes are found in the supra-nuclear cytoplasm. Bar = 2 μ m.

(Zamberletti *et al.* 1983). The finding that subsequent fertility of the patient is often not correlated with the previous extent of deciliation due to hydrosalpinx might suggest that other factors in addition to the recuperative activity of the tubal epithelium are affecting the normal function of the fallopian tube (Fedele *et al.* 1984; Afzelius *et al.* 1978).

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Inflammatory cell infiltration and survival in squamous cell carcinoma of the vulva

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Summary. The prognostic significance of tumour infiltration by inflammatory cells in squamous cell carcinoma of the vulva was examined in a cohort of 34 patients surviving without recurrence for at least 5 years after radical surgery, and a comparative cohort of 35 patients who died of their diseases. Overall, heavy inflammatory infiltration correlated with a good prognosis and light infiltration with a poor one, independent of other indices such as differentiation, tumour size and nodal status. IgA-containing cell infiltration also correlated with a good prognosis but the presence of IgA-containing cells did not alone account for all the inflammation in the good prognosis group. An immunological response to the tumour may be influencing prognosis. At a practical level, the extent of inflammation appears, at least in this material, to be as useful a prognostic index as many more conventional ones.

The assessment of prognosis for patients with ordinary squamous cell carcinoma of the vulva remains difficult. In some series prognosis is said to be influenced by tumour differentiation (Way 1960; Monaghan 1985) while in others, tumour differentiation does not appear to be an important prognostic factor (Donaldson *et al.* 1981; Iversen *et al.* 1981; Andreasson *et al.* 1982). Overall tumour size is important, larger tumours

tending to have a poorer prognosis (Franklin & Rutledge 1971), but in some patients very tiny carcinomas may present at a stage where groin node metastases have already occurred (Hacker *et al.* 1983). Metastatic deposits in the inguinal lymph nodes are closely related to ultimate outcome (Morley 1976; Benedet *et al.* 1979; Monaghan 1985), but nodal status can only be determined from a histological examination of the resection specimen, and is thus of no use in planning a pre-operative management strategy.

The infiltration of a variety of tumours by mononuclear cells has for many years been taken as an indication of host immune response to tumour cells. Attempts have therefore been made to correlate the degree of inflammatory infiltration with prognosis in a variety of clinical settings. Positive correlations of enhanced survival in the presence of a vigorous 'host response' have been reported for several tumours including gastric carcinoma (Black *et al.* 1956), colonic carcinoma (Murray *et al.* 1975) and in squamous cell carcinomas of the throat (Bennett *et al.* 1971) and cervix uteri (van Nagell *et al.* 1978). The general concept of human

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tumour immunity has been reviewed by Cochran (1978).

Vulvar carcinoma has been noted in immunosuppressed patients (Jimerson & Merrill 1970; Choo & Morley 1980), and several authors have commented on the degree of inflammatory infiltration in the tumours in their series. Gosling *et al.* (1961) in a series of 133 patients noted significantly more 5-year survivors when the primary tumour was markedly infiltrated by lymphocytes and plasma cells, and Andreasson *et al.* (1982) in a series of 147 patients reached a similar conclusion. In the more recent series reported by Hacker *et al.* (1984) and Boyce *et al.* (1985) no such effect was demonstrated.

Given the conspicuous nature of the mononuclear inflammatory infiltrate in many patients with vulvar carcinoma, the confusion in the literature and the need to learn more about prognostic indices in the disease, this retrospective study of inflammation in vulvar carcinoma as related to prognosis was undertaken.

Patients and methods

Selection of material

Using a computerized database the records of 267 patients with squamous cell carcinoma of vulva referred to the Regional Department of Gynaecological Oncology, Queen Elizabeth Hospital (QEH), Gateshead, between July 1974 and August 1986 were reviewed. Of these 267 patients 97 were referred up to August 1981. The number of referrals steadily increased during the later part of this study. Only 129 patients had been followed up for at least 5 years and were eligible for this study. A further 60 patients were excluded from the analysis for the following reasons: 22 were operated on at other hospitals and not followed up at QEH; 10 had histological findings showing malignant melanoma, basal cell carcinoma and not squamous cell carcinoma; 18 had incomplete data because tissue samples were missing, damaged, or inadequate for study; six had a recurrence and the biopsy suggested a second primary tumour; and four died and the cause of death was uncertain. Two cohorts remained in the study: (i) The surviving group comprising 34 patients treated by radical vulvectomy with bilateral groin node dissection, all of whom survived a minimum of 5 years; tumour recurrence has not been observed in this time. (ii) The dying group comprising 35 patients

referred for radical surgery who either died of vulvar carcinoma or developed a histologically proven recurrence within the 5-year period.

From the hospital records the patients' ages at diagnosis were established along with the site and the greatest diameter of their tumours as recorded in the operation notes. From the histological slides the diagnosis of squamous cell carcinoma was confirmed, and the tumours were classified according to conventional histopathological criteria as well-differentiated, moderately well-differentiated or poorly differentiated. A category of basaloid growths was considered separately. The presence or absence of groin node metastases was confirmed.

A single representative block was selected for each patient. These usually included the tumour/skin interface and wherever possible the full thickness of the tumour. From these blocks, which had originally been fixed in Susa, multiple spare sections were prepared.

Scoring of slides: overall inflammation index

One of the spare sections from each case was stained by haematoxylin and eosin, and the slides were randomized. Following a preliminary screening to establish the range of infiltration by mononuclear cells to be expected in the material, the degree of infiltration in each case was assessed on a scale of 1 to 3, corresponding to a mild, moderate or marked involvement. Separate scores were produced for the tumour edge and for the tumour stroma to try to account for the variable density of different growths. At this stage no attempt was made to discriminate between cell types in the infiltrate. All mononuclear cells were counted. Consistency of assessment was checked by random reassessment of previously assessed slides. For purposes of analysis the two scores were added, since tumour edge scores paralleled tumour stroma scores and there were no consistently significant differences. This gave an overall inflammation index which ranged between 2 (minimal inflammation) and 6 (marked inflammation).

Scoring of slides: immunoperoxidase studies

Spare sections from each case were stained by a three-stage immunoperoxidase (PAP) technique as described by Sternberger (1979), using commercially obtained antisera to IgA, IgE, IgG and IgM heavy chains, κ and λ light chains

Table 1. Distribution of 'surviving' and 'dying' groups of patients with squamous cell carcinoma of the vulva by tumour differentiation, size, and groin node status

Variable	'Surviving' group	'Dying' group
Degree of tumour differentiation		
Good	11	3
Moderate	7	5
Poor	14	22
Basaloid	2	5
Tumour size*		
<25 mm	15	5
25-50 mm	15	11
>50 mm	4	17
Nodal status†		
Negative	31	6
Positive	3	20

*Maximum diameter recorded at operation, the size of two tumours in the 'dying' group was not recorded.

†Histological evidence of nodal involvement was not available in nine patients in the 'dying' group.

Table 2. Distribution of patients in the two groups by overall inflammation index of tumour

Patient group	Inflammation index				
	2	3	4	5	6
'Surviving' (n=34)	2	3	11	11	7
'Dying' (n=35)	18	10	4	3	0

and muramidase (Dako, Denmark). The sections were trypsinized before application of the primary antisera, and the optimal dilution of each antibody was established by titration. The chromogen used was diaminobenzidine and colour development was enhanced by the application of a copper sulphate solution (Lauder *et al.* 1984). Sections were counterstained with haematoxylin and randomized for assessment.

The slides were randomized in such a way that the analysis of each batch of slides stained by a single antiserum was completed before the next was started. As previously, a preliminary screening established the range of positivity to be expected within a particular batch of slides; and then the numbers of positive cells infiltrating at tumour edge and stroma were assessed independently on a scale of 1 to 3. Consistency of assessment was checked by the random testing of previously assessed slides.

As before, tumour edge scores paralleled

those of the stroma, with no consistent patterns emerging, and therefore edge and stroma were added together to give an overall index for each of the separate antisera.

Results

Initial comparison of 'surviving' and 'dying' groups

The mean age at presentation in the 'surviving' group was 67.3 years and in the 'dying' group 68.8 years. This small difference is not statistically significant. The age distributions at presentation in the two cohorts were virtually identical, with most patients over 70 years of age at diagnosis and only two or three women under 50 years of age.

Table 1 shows the distribution of the patients in the two groups by tumour differentiation, tumour size and the presence or absence of metastatic deposits in groin lymph nodes. Seven of the tumours in the series had a basaloid growth pattern and we have segregated these cases from the 'ordinary' squamous cell carcinomas for the purpose of statistical analysis. A full range of differentiation patterns was evident but there was a significant preponderance of poorly differentiated lesions in the 'dying' group ($\chi^2=6.6$, $P<0.05$). Looking for a trend in prognosis related to degree of differentiation, a more significant difference was observed ($\chi^2=6.6$, $P<0.01$). When tumour size was assessed, prognosis was worse in large tumours than in small. Statistical analysis taking into account the trend that the proportion of treatment failures increases as the size of the tumour at presentation increases showed a significant difference ($\chi^2=12.9$, $P<0.001$). Nodal status was obviously correlated with outcome ($\chi^2=28.9$, $P<0.001$).

Prognosis in these patients was thus related to the degree of differentiation and size of their tumours and to the presence or absence of nodal involvement.

Inflammatory cell infiltration in 'surviving' and 'dying' groups

Table 2 shows the number of cases in the 'surviving' and 'dying' groups according to the overall inflammatory indices of their tumours. There was a significant predominance of heavily infiltrated tumours in the 'surviving' group and an equally striking or significant preponderance of

tumours lacking inflammatory infiltration in the 'dying' group (Wilcoxon rank-sum test statistic $Z=5.3 \sim N(0,1)$ $P<0.001$).

Tables 3 and 4 show the number of cases in 'surviving' and 'dying' groups expressed according to inflammatory index in relation respectively to tumour differentiation and tumour size. Table 3 shows that in all four differentiation categories 'surviving' patients tended to have higher inflammatory indices than those 'dying'. Some of the differentiation classes contained relatively small numbers of tumours but applying the Wilcoxon rank test to the group with the largest number of tumours, poorly differentiated carcinomas, the difference between 'surviving' and 'dying' groups was significant ($Z=4.2 \sim N(0,1)$ $P<0.001$). Therefore, even controlling for differentiation, inflammatory infiltration was a powerful prognostic indicator in this study. Similarly Table 4 shows that the influence of inflammation on prognosis was independent of tumour size as a risk factor. Formal statistical evaluation of the tumours in the medium size category (25–50 mm) by the Wilcoxon rank test again shows a significant difference ($Z=3.5 \sim N(0,1)$ $P<0.001$). The prognostic value of inflammatory infiltration was also apparently independent of nodal status, at least

Table 3. Number of cases expressed as the ratio of surviving/dying patients in each category of overall inflammation index by tumour differentiation

Degree of tumour differentiation	Inflammation index				
	2	3	4	5	6
Good	1/2	1/0	3/1	3/0	3/0
Mod. rate	1/1	2/2	1/1	1/1	2/0
Poor	0/11	0/7	6/2	6/2	2/0
Basaloid	0/4	0/1	1/0	1/0	0/0

Table 4. Number of cases expressed as the ratio of surviving/dying* patients in each category of overall inflammation index by tumour size

Tumour size	Inflammation index				
	2	3	4	5	6
<25 mm	2/3	1/1	4/1	5/0	3/0
25–50 mm	0/3	2/6	5/2	4/0	4/0
>50 mm	0/12	2/2	2/1	2/2	0/0

*For two of the dying group overall size was not known.

in node-positive patients where there was a reasonable number of patients in both 'surviving' and 'dying' groups ($Z=2.5 \sim N(0,1)$ $P<0.05$).

Infiltration by individual cell types—immunoperoxidase study

Table 5 shows the number of cases in 'surviving' and 'dying' groups expressed according to the various inflammatory indices ascribed to them after assessment of the immunoperoxidase-stained slides. It is to be stressed that the indices were arrived at as an estimate of relative numbers of positive cells present, uninfluenced by background staining or the intensity of the final reaction product. Their absolute number was not estimated, but it was noted that as a rule lymphoid cells containing κ and λ light chains and IgG were plentiful in the tissues, IgA- and IgM-containing lymphoid cells and muramidase-containing macrophages less plentiful, and IgE-containing lymphoid cells rare. No formal attempt was made to compare the intensity of infiltration by one type of cell with infiltration by another in particular tumours, the object being to establish whether the relative frequency of any particular cell type was associated with a particular prognostic group.

Table 5 shows that as far as cells containing κ and λ light chain, those containing IgG, IgM and IgE heavy chain and those containing muramidase are concerned, there was no particular association with either the 'surviving' or 'dying' group of patients; statistical analysis confirms that impression. In the case of IgA-containing cells, however, the intensity of infiltration was significantly less in the 'dying' group and greater in the 'surviving' group (Wilcoxon rank test, $Z=3.5 \sim N(0,1)$ $P<0.001$). As might be expected, the IgA inflammation index correlated closely with the overall inflammation index estimated from haematoxylin and eosin stained slides.

Discussion

In this series of patients, survival was influenced by tumour differentiation, tumour size and the absence of metastatic diseases in regional lymph glands, but a considerable number of patients did badly despite having favourable prognostic indices and some did better than anticipated.

Thus there appear to be other factors influencing prognosis.

Infiltration of tumours by mononuclear cells assessed on routine haematoxylin and eosin stained sections appears to be a useful prognostic indicator. Heavily infiltrated tumours had a good prognosis and those which were lightly infiltrated a poorer prognosis, independent of the other prognostic factors assessed. It is clearly not simply related to ulceration or infection since blocks were selected only from sites where such changes were inconspicuous although ulcerated tumours were not excluded from this series.

It is difficult to be certain that previous biopsy did not affect the inflammatory infiltrate but, where a biopsy had been taken, there was no evidence of previous surgery or ulceration in the blocks examined. Conventionally the mononuclear inflammatory infiltrate has been regarded as being morphological evidence of an immune response to tumours. If, as seems likely, the mononuclear cells present in the vulval carcinomas in this series are involved in a host response, what do immunoperoxidase studies tell us of its nature?

Muramidase (lysozyme) is an enzyme found in large quantities in a variety of cell types (Mason & Taylor 1975). It is particularly conspicuous in cells of the myeloid series and in cells of the monocytic series. In the context of the present study the muramidase-positive mononuclear cells whose relative numbers were assessed represent the population of tissue macrophages. Although their numbers were variable and in some cases quite large, no correlation was seen between density of infiltrate and prognosis. Thus despite the accepted role of macrophages in the immune response, infiltration by this series of cells does not seem to contribute to the excess of inflammation seen in tumours of good prognosis in this study.

Immunoglobulins are secreted by cells of the B-lymphocyte series and thus the presence of cytoplasmic immunoglobulin components (κ or λ light chains or IgA, IgG, IgM or IgE heavy chains) implies a degree of maturation along the pathway of B-lymphocyte differentiation. No significant difference was observed between 'surviving' and 'dying' groups as far as infiltration by κ and λ light chain positive cells was concerned. This implies that the excess of inflammation seen in the 'surviving' group is not a consequence of a general increase in the

Table 5. Number of cases expressed as the ratio of surviving/dying* patients in each category of inflammation index for the several types of antisera used

Antiserum	Inflammation index				
	2	3	4	5	6
κ -positive cells	5/10	8/9	10/5	5/6	6/4
λ -positive cells	7/9	6/8	13/7	4/5	4/4
IgG-positive cells	12/10	6/9	6/8	5/4	5/4
IgM-positive cells	25/25	6/7	1/1	1/1	1/1
IgE-positive cells	34/34	—	—	—	—
Muramidase-positive					
macrophages	19/15	3/8	3/5	5/3	4/3
IgA-positive cells	3/19	1/6	19/3	5/1	6/6

*For one dying patient material was not available for all special staining procedures.

numbers of infiltrating B-cells. Nor was there any difference in infiltration by numbers of cells containing IgG, IgM or IgE. Local production of these immunoglobulin groups would thus appear to be unimportant in prognosis. Somewhat unexpectedly a marked excess of IgA-containing cells was seen in the 'surviving' group and a paucity of such cells in the 'dying' group. The significance of this observation is not clear. It is, however, noteworthy that elevated serum IgA levels have been reported in patients with squamous carcinoma (in the nasopharynx for example, Wara *et al.* 1975), and IgA has been found in some extracts made from tumours (Cochran 1978). Local IgA production has also been reported as important to the function of T-lymphocytes (Rosenthal 1980; Rheinherz & Schlossman 1980). Whatever the local role of the IgA-containing cells, their absolute numbers were small and insufficient to account for the excess of inflammation seen in the 'surviving' group.

Vulvar carcinoma remains a difficult disease to treat and modern management is increasingly demanding a better pre-operative assessment of patients in order that treatment can be individualized (Hacker *et al.* 1984). Overall tumour size is clearly of prognostic importance: in practical management terms a tumour diameter of 40 mm has been recently suggested to represent a watershed at which prognosis alters and some management strategies must be changed (Andreasson *et al.* 1982; Monaghan & Hammond 1984). We would suggest that the presence or absence of a dense lymphoid infil-

trate within the tumour may offer a similar rule of thumb for the clinician.

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Successful pregnancy after selective embolization of a post-molar vascular malformation. Case report

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Case report

A 27-year-old married nullipara stopped her oral contraceptive in May 1982 and immediately became pregnant. When she was referred for hospital booking in October 1982, after 14 weeks amenorrhoea, it was noted that her uterus was 4 weeks larger than expected. An ultrasound scan showed 'a bulky uterus with molar pattern and no fetal parts. Both ovaries contain large cysts—presumably theca lutein'. Her chest X-ray, and thyroid and liver function tests were normal. The titre of urinary hCG was 1/5000 equivalent to 2.5 million i.u./24 h.

The hydatidiform mole was evacuated by suction under general anaesthesia 48 h later and 60 g of grape-like material was sent to the pathologist who reported 'a typical complete hydatidiform mole showing moderate hyperplasia and some pleomorphism with mitotic activity'. She was discharged from hospital the next day and her case registered with the Trophoblastic Screening Centre in Sheffield, who undertook to monitor weekly samples of urine for hCG.

Bleeding continued for the next 6 weeks and urinary hCG remained elevated at 45 000 i.u./24 h. On 23 December 1982 her uterus was explored under general anaesthesia and a further 13 g of molar tissue was obtained. The pathologist reported 'trophoblastic hyperplasia, no myometrium; a complete mole'. Follow-up continued and as the hCG levels remained persistently elevated she was admitted to the

Sheffield unit five times between January and June 1983 for low dose methotrexate/folinic acid therapy. In June 1983 she started taking a combined oral contraceptive pill, given in a cyclical pattern. She subsequently had monthly withdrawal bleeding with moderate losses. In December 1983 she discontinued her pill and 2 weeks later had a flooding vaginal haemorrhage and was admitted to hospital as an emergency. She was shocked, with a haemoglobin level of 7.1 g/dl, and was given a total of 10 units of blood. She was treated with intramuscular progesterone daily for 1 week and her pill restarted to give long-term control. On discharge home her Hb was 11.5 g/dl.

A pelvic angiogram was arranged and was reported as follows: 'uterus enlarged and shows marked vascularity. Early A-V shunting of blood occurs. Both internal iliac arteries are enlarged. Appearance represents persistent hypervascularity following a successfully treated mole'. Hysteroscopy showed no evidence of endometrial lesions. Following discussion with the patient, the plan was to continue with oral contraception until September 1984, and then she would try for a pregnancy.

In September 1984 the 'pill' was discontinued, and as the first cycle was anovulatory clomiphene was given to induce ovulation. Between October 1984 and January 1985 she was admitted to hospital five times with heavy bleeding and required further blood transfusions on two occasions. From January 1985 a 50 µg 'pill' was prescribed and this controlled the severe menorrhagia. At this time a CT scan showed 'A bulky uterus which is surrounded by a hypervascular mesh of small vessels. Both internal iliac arteries are enlarged. No enlarged lymph nodes or anything to suggest residual trophoblastic tumour'. hCG levels were normal.

The patient was adamant that she wanted a child and wished to avoid any suggestion of hys-

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terectomy. Consideration was therefore given to ligation of the internal iliac arteries, but after consultation with the radiologist (H.W.L.) it was decided that selective embolization of these vessels be undertaken in an attempt to control the heavy bleeding.

Selective embolization

Angiography showed a large malformation composed of medium-sized vessels with rapid venous shunting into very enlarged (1.5 cm diameter) draining veins, and pooling of contrast medium in vascular spaces. The abnormal vessels involved the uterine body and fundus with extension into the adnexa particularly on the left side (Fig. 1). There were bilateral enlarged uterine vessels and uterine veins reflecting the extensive spread of tumour. The malformation was embolized in a two-stage procedure.

At stage I the anterior branch of the left internal iliac artery was catheterized and the internal pudendal artery was blocked in a distal position beyond the uterine vessels with a Gianturco metal coil (Cook Europe), so that perfusion would be maintained via the collateral circulation from the external pudendal to the terminal branches, thus ensuring the preservation of the blood supply to the bladder base and external genitalia. Two large uterine vessels were embolized by injection of small fragments of Sterispon and polyvinyl alcohol (Ivalon) suspended in a normal saline and contrast medium (Iohexol 350) in equal quantities. Finally a small spring



Fig. 1. Venous phase of aortogram demonstrating uterine vascular malformation and enlarged draining veins, 1983.

coil was placed in each 'feeding vessel': those un-named arteries which deliver blood to the A-V malformation.

At stage II, 10 weeks later, two branches of the posterior segment of the left internal iliac were embolized in the same way and the anterior branch of the right internal iliac was catheterized and two large uterine vessels were embolized, utilizing fragments of Sterispon, polyvinyl alcohol and five small Gianturco coils (Fig. 2). After this procedure residual perfusion was present in the left adnexa only (Fig. 3). Recovery from the procedure was uncomplicated.

Results

After 2 months she was given a 5-day course of clomiphene, 50 mg daily. As judged from a plasma progesterone estimation, there was no ovulatory response in the first cycle. A second course of clomiphene, 100 mg daily, was given and a good ovulatory response occurred (progesterone 41.2 nmol/l). She missed her next period and when 2 weeks overdue had a positive pregnancy test, her hCG level rising to 60 000 i.u./l.

Her pregnancy progressed quite normally and an ultrasound scan showed a normal singleton fetus with normal placentation. At 30 weeks gestation she was admitted to hospital as an emergency, complaining of abdominal pain and backache. There had been a history of a 'flu-like' illness 2 weeks previously. She was pale, with a pulse rate of 120 beats/min, blood pressure 110/70 mmHg and a fetal CTG showed decelerations down to 70 beats/min. An emergency caesarean section was done through a vascular lower segment and a female child weighing 1389 g was born. The blood loss was 1200 ml and she was given two units of whole blood. The placenta which was small (340 g) showed areas of 'septic' infarction on histology. No organisms were grown from it. The baby developed respiratory distress and required ventilatory support for 3 days. *Listeria* was cultured from the baby's blood but there was no evidence of systemic infection. Seven weeks after birth the baby weighed 2.27 kg and was breast-feeding; the mother was fit and well.

Discussion

Life-threatening blood loss may be associated with postpartum or postoperative haemorrhage,



Fig. 2. Stage II: Arterial phase of right internal iliac. Post-embolization of the left. Note multiple coils in left internal iliac branches, and increase in size in 3 years.

or pelvic cancer. Previously the surgeon had to resort to ligation of the uterine iliac arteries to control such bleeding: a major procedure often carried out on a shocked and already compromised patient. Transarterial embolization has proved to be a safer alternative, with the added advantage that it may be performed under local anaesthesia (Vujic *et al.* 1986; Pais *et al.* 1980; Adam *et al.* 1986; Rosenthal & Colapinto 1985; Greenwood *et al.* 1987). The use of pelvic angiography in the investigation of malignant trophoblastic disease has shown that despite adequate therapy arterio-venous abnormalities may persist in the pelvis (Cockshott &

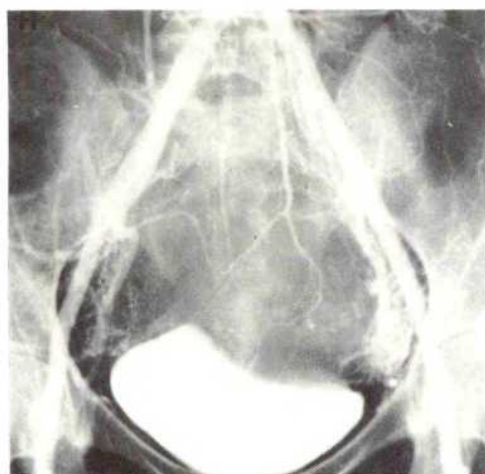


Fig. 3. Post-embolization, right and left. Residual arterial perfusion in left adnexum region only.

Hendrickse 1967; Stern *et al.* 1968). Post-molar life-threatening haemorrhage which was successfully treated by non-selective bilateral internal iliac embolization has been reported by Vujic *et al.* (1986). Our patient was a young woman, wanting a successful pregnancy, whose fertility was compromised by a post-molar pelvic vascular abnormality which was producing excessive vaginal bleeding and the need for recurrent blood transfusion. The bleeding could only be controlled by prescribing a high-oestrogen contraceptive which denied conception and increased the risk of thromboembolism and cardiovascular complications.

The condition is rare and although bilateral internal iliac artery ligation was considered as an acceptable form of treatment and could have been followed by a successful pregnancy, the patient would have been subjected to the dangers and inconvenience of a laparotomy and a general anaesthetic. At the time (1984/85) the skills of interventional radiology in percutaneous transarterial embolism were being developed, and this method was considered to be a much safer and less stressing alternative for our patient (Reidy 1987).

The advantages of an outpatient procedure using local anaesthetic over open surgery are self-evident.

A two-stage procedure for embolization was chosen in order to ascertain that the left side, which was the most involved in the vascular abnormality (Fig. 1), had been deprived of its feeding vessels for at least 4 weeks and had not developed new sources of supply, before performing the less extensive procedure on the right side which was slightly less involved. The two-stage procedure is recommended to establish that good devascularization of the major malformation has been successfully achieved before completing the treatment. Selective embolization is more likely to result in adequate pelvic vascularity for a future pregnancy than arterial ligation and should be considered whenever pelvic A-V malformation is associated with severe uterine haemorrhage.

In our patient successful embolization allowed the resumption of normal menstruation and later a conception that resulted in the birth of a surviving child. It could be asked whether the placental infarction was attributable to the embolization. The history suggests a probable infective cause, possibly due to listeriosis, which in turn may have been responsible for the pre-

mature onset of labour. The baby's weight was on the 50th centile for gestation, suggesting that intrauterine growth had not been prejudiced by the embolization. To our knowledge this is the first report of a successful pregnancy following embolization for this condition.

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Successful pregnancy after radical surgery for diethylstilboestrol (DES)-related vaginal adenocarcinoma. Case report

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Case report

This woman had presented at the age of 24 in 1983 with a clear-cell vaginal adenocarcinoma. She had been exposed '*in utero*' to maternally ingested diethylstilboestrol taken from the 9th to 16th week of pregnancy for threatened miscarriage. Treatment consisted of removal of the vagina and vaginal portion of the cervix together with the adjacent segment of the bladder and urethral wall. The pouch of Douglas was opened so that the supravaginal cervix could be divided above the vaginal fornices. This perineal procedure was accompanied by bilateral extra-peritoneal pelvic lymphadenectomy. The cavity was packed using Whitehead's varnish, and 1 week later reconstruction of the vagina was performed with a splitskin graft taken from the thigh (McIndoe-Jayes technique). The apex of the graft was fenestrated for apposition to the supravaginal cervix (Hudson *et al.* 1983).

Following surgery, the patient was kept under regular surveillance without evidence of recurrence or reactivation of the malignancy.

Some contraction of the neo-vagina occurred and the donor site was afflicted with a 'flare-up' of pre-existing psoriasis. It was extremely difficult to identify the site of the 'cervix' but regular menstruation occurred and it was possible to locate a 'pinpoint' aperture communicating with the uterus.

A period of primary infertility ensued, to

which a male factor was contributory. At a late stage diagnostic curettage was carried out. It was eventually decided that the couple might be suitable for gamete intrafallopian transfer (GIFT). However, conception occurred in 1986 without further intervention.

The pregnancy

Pregnancy proceeded uneventfully until 36 weeks. Elective caesarean section had been determined as the method of delivery. Consideration was given to measures prophylactic against mid-trimester miscarriage. It was recognized that the uterine abnormalities associated with maternal DES ingestion might, if present, themselves predispose to early pregnancy loss. The complete absence of the vaginal portion of the cervix precluded cerclage except by the abdominal route and a decision was taken by agreement with the patient to await events. At 36 weeks spontaneous premature labour began and a female infant was born by emergency caesarean section; the baby thrived.

At operation the uterus was found to be of normal shape, as were the adnexa, and there were no metastases noted.

Discussion

Loss of reproductive capability is a particularly distressing side-effect of the radical management of lower genital tract malignancy in adolescents and women. In DES-related cases it provides additional psychological trauma for the patient's mother, who will already be subject to feelings of guilt.

The potential for preservation of reproductive function in selected cases treated by carefully tailored radical surgery has now been realized

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with a successful spontaneously conceived pregnancy.

The risks of suboptimal therapy with the same intent have been documented (Herbst & Anderson 1981). It is important that chances of survival should not be compromised by local excision without an adequate margin of normal tissue in three dimensions. Because of a propensity for lymphatic metastasis the additional need for lymphadenectomy is emphasized.

Acknowledgment

This patient was managed and followed up in the Combined Gynaecological Oncology Service at

Westmead Hospital, New South Wales, Australia.

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Angiomyolipoma of the vagina—an uncommon tumour. Case report

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Case report

A 46-year-old Indian woman presented to the Department of Obstetrics and Gynaecology, University Hospital, Kuala Lumpur, in September 1986, with a complaint of progressive lower abdominal swelling of 3 months duration, but without abdominal pain. Her menses had been normal and there was no history of disturbance in micturition. On pelvic examination, the cervix was thought to be rather soft and ballooned out. The external os was not visible on speculum examination. The uterus was felt to be large, with the fundus almost reaching the level of the umbilicus. A pre-operative diagnosis of uterine myoma was made.

Pre-operative intravenous pyelogram showed lateral displacement of the pelvic portion of the ureters with minimal hydro-ureter on the right side. There were no radiolucent areas in either kidney to suggest the presence of renal tumour. The patient also revealed no features suggestive of tuberous sclerosis.

At laparotomy, the bladder was found to be displaced superiorly. In between the bladder and the normal-size uterus was a solitary large tumour measuring 20×12×10 cm. This tumour appeared to arise from the anterior wall of the vagina (Fig. 1). It was densely adherent to the bladder and was dissected out with difficulty. A total hysterectomy, bilateral salpingectomy and complete excision of the mass was performed. Intra-operative diagnosis of a sarcoma was made.

The tumour which arose from the vaginal wall below the mucosa weighed 900 g. Its cut surface revealed solid yellowish fleshy tissue, which was relatively soft in consistency. It was composed of a mixture of anastomosing clusters of thick-walled blood vessels, elongated spindle-shaped, smooth muscle cells and adipose tissue of the adult type (Figs 2 & 3). There were no pleomorphic or immature features present. Sarcomatous components were not seen. These features indicate a diagnosis of angiomyolipoma.

The patient made an uneventful recovery after the operation, and was discharged 1 week later. She was well when last seen at the follow-up clinic 4 months later.

Discussion

Angiomyolipoma was first documented by Bourneville in 1880 (Ma & Chan 1974). In early years it was given various other names such as 'myoangioliipoma' by Bartsh in 1900 (Tweeddale *et al.* 1955), 'benign arterioleiomyoma' by Gordon *et al.* (1939) and 'neurilemmoblastosis' by Inglis (1950). However, it is most appropriately called by its present descriptive name, 'angiomyolipoma', the term first used in 1951 by Morgan and his associates (Hadju & Foote 1969) because of the presence of the three intimately mixed but distinctive components in the tumour: the spindle-shaped smooth muscle cells which are believed by some authors to arise from the wall of the arterioles (Inglis 1960), clusters of anastomosing thick-walled blood vessels which resemble small and medium-sized arteries and adipose tissue of adult type.

The histopathology of angiomyolipoma has been well studied, based primarily on the renal tumour. The histology of these tumours is usually constant although the proportion of the three components may show slight variation from tumour to tumour. The microscopic findings of the tumour removed from the vagina of

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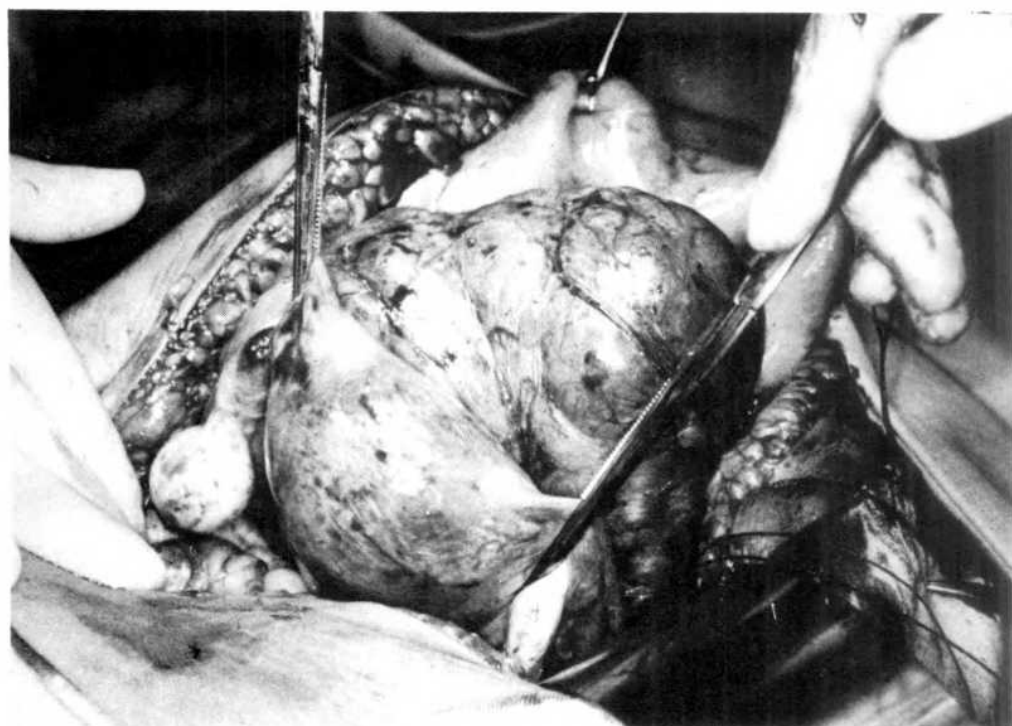


Fig. 1. Tumour, measuring $20 \times 12 \times 10$ cm, arising from the anterior wall of the vagina situated between the urinary bladder and uterus.

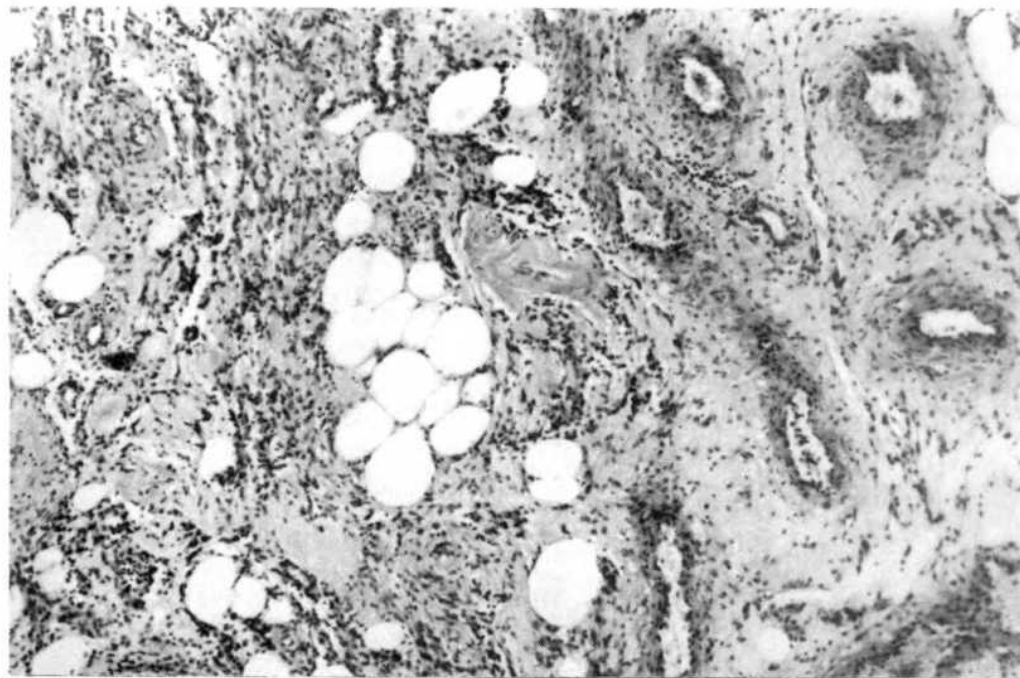


Fig. 2. Cluster of thick-walled blood vessels in the tumour. Haematoxylin & eosin $\times 20$.

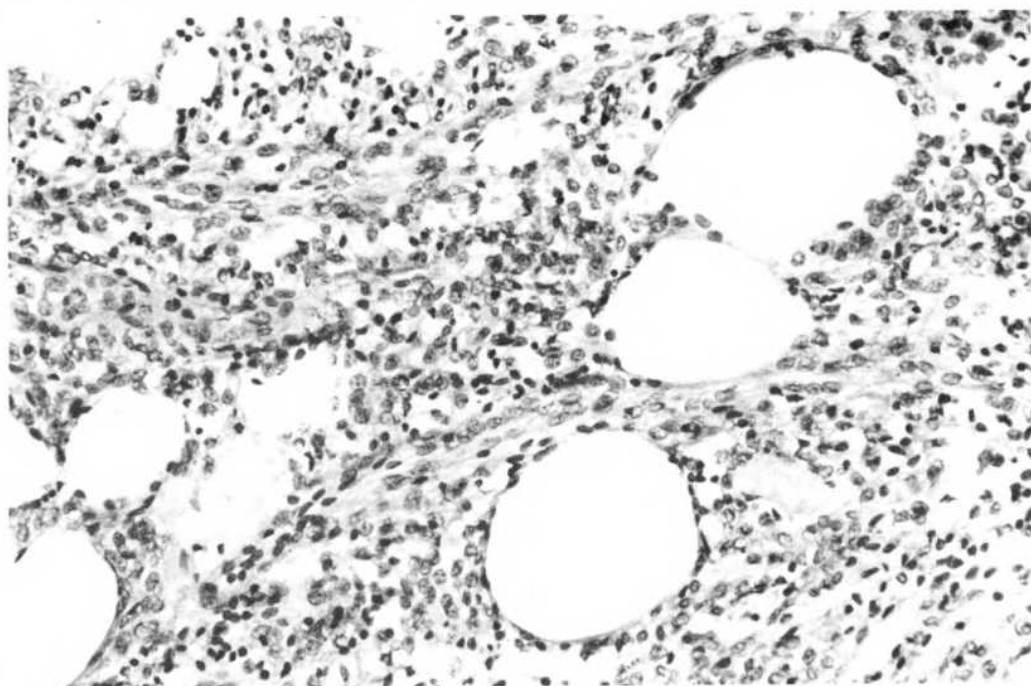


Fig. 3. Spindle-shaped smooth muscle cells and adult fat cells. Haematoxylin & eosin $\times 200$.

our patient were found to be in every aspect similar to the well-documented renal angiomyolipoma. However, the histological appearance of angiomyolipoma is in turn indistinguishable from lipoleiomyoma (Demopoulos *et al.* 1973; Hendrickson & Kempson 1980; Kempson & Hendrickson 1987), a rare lipomatous tumour of the uterine corpus. The latter may be seen in the uterine cervix, but the vagina is an exceedingly uncommon site. In view of the similarity between angiomyolipoma and lipoleiomyoma, it had been suggested that both these lesions share a similar origin (Demopoulos *et al.* 1973). In some ways, the appearance of this tumour also resembles angioleiomyoma, another variant of leiomyoma.

The histogenesis of angiomyolipoma is still in doubt. It is classified as a hamartoma by some authors (Moolten 1942; McCullough *et al.* 1971), whereas it is regarded by others as a neoplastic lesion (Tweeddale *et al.* 1955), whereby the smooth muscle cell is thought to be the neoplastic component. The distinctive presence of mature lipocyte and thick-walled vessels is still an enigma. Nonetheless angiomyolipoma is a benign lesion and it is widely accepted that malignant change is extremely rare (Ma & Chan 1974). The few reports which suggest sarcomatous lesions are thought to be not well sub-

stantiated (Farrow *et al.* 1968). Misdiagnosis of malignancy may occur (Ma & Chan 1974; McCullough *et al.* 1971). Some authors favour a multicentric origin of this lesion when more than one nodule is present in an organ or within the regional lymph node (Busch *et al.* 1976). There are very few reports of angiomyolipoma occurring in extra-renal organs. It is interesting to note that all these patients had tuberous sclerosis and often had associated renal angiomyolipomas (Tweeddale *et al.* 1955). Studies show that only 50% of renal angiomyolipoma were diagnosed in tuberous sclerosis patients even though 80% of tuberous sclerosis patients were found to have angiomyolipoma (McCullough *et al.* 1971; Walker *et al.* 1976). Our patient did not have features suggestive of tuberous sclerosis and there was no evidence of renal tumour in intravenous pyelogram.

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SHORT COMMUNICATIONS

Breast sensitivity and lactational amenorrhoea

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Ovulation precedes first menses in about 20% of lactating women (Howie *et al.* 1982). In one series of 12 breast-feeding women, two conceived without menstruating (McNeilly *et al.* 1983). A simple method of predicting ovulation during lactation could be helpful to women unable or unwilling to use contraception, particularly in developing countries. After delivery there is a sharp increase in breast sensitivity (Robinson & Short 1977), probably due to falling steroid levels (West & McNeilly 1979), but it is not known whether breast sensitivity decreases again when ovarian activity resumes in late lactation. Increased oestrogen production usually precedes the first menses by several weeks (McNeilly *et al.* 1982), and decreasing breast sensitivity might therefore predict the end of lactational infertility.

Patients and methods

Twelve women, recruited at the antenatal clinic, gave fully informed consent. All were married and had had regular menses before conceiving. All had vaginal deliveries of normal babies at term. Of the six who breast-fed, three were multiparous and three primiparous, five were white European and one was Indian, and their mean age was 29 years (range 24-39). The six who bottle-fed were white: two were multiparous and four primiparous and their mean age was 28.5 years (range 23-37). Subjects were visited in their homes twice-weekly by one investigator who measured breast sensitivity.

Breast sensitivity measurements

Two-point discrimination was measured by bringing together the points of a pair of geometric dividers, initially widely separated, until two distinct sensations were no longer felt, and the distance between the points was recorded. The medial and lateral half of each breast was tested separately on each occasion.

Touch sensitivity was measured with a Semmes-Weinstein pressure aesthesiometer (Shaw Laboratories, Inc., USA), consisting of 20 nylon monofilaments of varying diameter (Hinde *et al.* 1963). Each filament was pressed against the skin until it bent slightly and the smallest size that could be felt was recorded. The nipple, areola and cutaneous tissue of each breast were tested separately on each occasion.

Urinary steroid measurements

Samples of early-morning urine were collected twice-weekly. Total oestrogens and pregnanediol were measured by radioimmunoassay and the concentration expressed in relation to creatinine concentration. Among lactating women, the frequency of feeding, particularly at night, was regularly noted. Among most subjects the study was terminated at the first menses, but observations were continued on four women who were breast-feeding until the second or third menses.

Results

The mean duration of amenorrhoea among breast-feeding women was 24.3 weeks (range 14-35), and among bottle-feeding women was 7.5 weeks (range 6-14). Among breast-feeding women the mean duration of observations was 30 weeks (range 18-40).

Urinary steroids

Among the lactating women, four showed luteal activity in the week before the first menses, with

peak urinary pregnanediol concentrations of 0.91, 1.53, 0.91 and 0.97 nmol/nmol creatinine, respectively. Each pregnanediol peak was associated with, or immediately preceded by, a peak of oestrogen excretion, of 19.8, 18.5, 14.8 and 15.2 nmol/nmol creatinine, respectively. The other two women had peak pregnanediol concentrations of only 0.20, and 0.26 before first menses: in one there was no oestrogen peak, but in the other a peak of oestrogen excretion preceded the first menses, reaching a level of 20.1 nmol/nmol creatinine.

Among the non-lactating women, one showed luteal activity before the first menses, with a peak urinary pregnanediol concentration of 0.85 nmol/nmol creatinine 1 week before the menses. Another had a peak pregnanediol level of 0.34 in the week before first menses, and another a peak of 0.42 nmol/nmol creatinine more than 2 weeks before the menses. The other three showed no increase in pregnanediol. Two non-lactating women showed peaks of oestrogen excretion (>20 nmol/nmol creatinine) in the fortnight before first menses.

Two-point discrimination

As oestrogen levels rose, three of the six lactating women showed a decrease in two-point discrimination (that is, an increase in the distance between the points that could be discriminated). However, in one subject, two-point discrimination improved as lactation progressed, and two other subjects showed a slight improvement also. In one subject two-point discrimination showed some promise as an indicator of increasing oestrogen levels (Fig. 1) but among the six lactating women the ranges of the readings were almost identical in early and late lactation. Among non-lactating women four showed a slight improvement in two-point discrimination but in this group also the ranges were similar in the early and late puerperium.

Touch sensitivity

In four lactating women the mean touch threshold rose as oestrogen levels increased, but in the other two the change was minimal. The rise in touch threshold was not an accurate predictor of the first menses: in one subject a greater increase occurred after first menses than before (Fig. 1). The ranges were similar in early and late lactation. Among non-lactating women only one

showed a rise in mean touch threshold, and in this group too the range was similar in the early and the late puerperium.

Discussion

During early lactation ovarian activity is suppressed completely (Glasier *et al.* 1984) and resumes between the third and fifth weeks postpartum (McNeilly *et al.* 1982), in association with a decline in suckling frequency (McNeilly *et al.* 1983) and with the introduction of supplementary feeding (Gross & Eastman 1983). All but one of our lactating women had stopped night feeds by the time of their first menses, and one did not menstruate until she had completely stopped breast-feeding. The first menses may follow anovulatory cycles, ovulatory cycles, or cycles in which the luteal phase is deficient (McNeilly *et al.* 1982, 1983): in our study, four of the six lactating women showed evidence of luteinization before first menses, though we could not assess whether the luteal phase was adequate.

Oestrogen can affect skin sensitivity in animals (Komisaruk *et al.* 1972). Robinson & Short (1977) showed that breast sensitivity increases among girls (but not among boys) at puberty, varies during the menstrual cycle and increases after delivery. We hypothesized that a decrease in breast sensitivity would occur as lactation progressed, and our study confirmed that touch sensitivity tends to decrease as lactation progresses, though two-point discrimination shows no consistent change. However, the change in touch sensitivity is unlikely to be of practical value as an indicator of returning fertility for several reasons. Firstly, the decrease in sensitivity is gradual, unlike the sudden increase in sensitivity at parturition. Secondly, the overall range of values for both touch threshold and two-point discrimination is similar in early and late lactation, and it was not possible to define a 'threshold' which would identify a sudden rise in oestrogen levels. Thirdly, not all subjects showed the decrease in sensitivity. Finally, it was disappointing that the decrease in sensitivity could be detected only by the aesthesiometer (a relatively sophisticated device), and not by the simpler and cheaper geometric dividers.

We conclude that although breast sensitivity decreases during lactation, the change is so gradual and so difficult to detect that it is of no

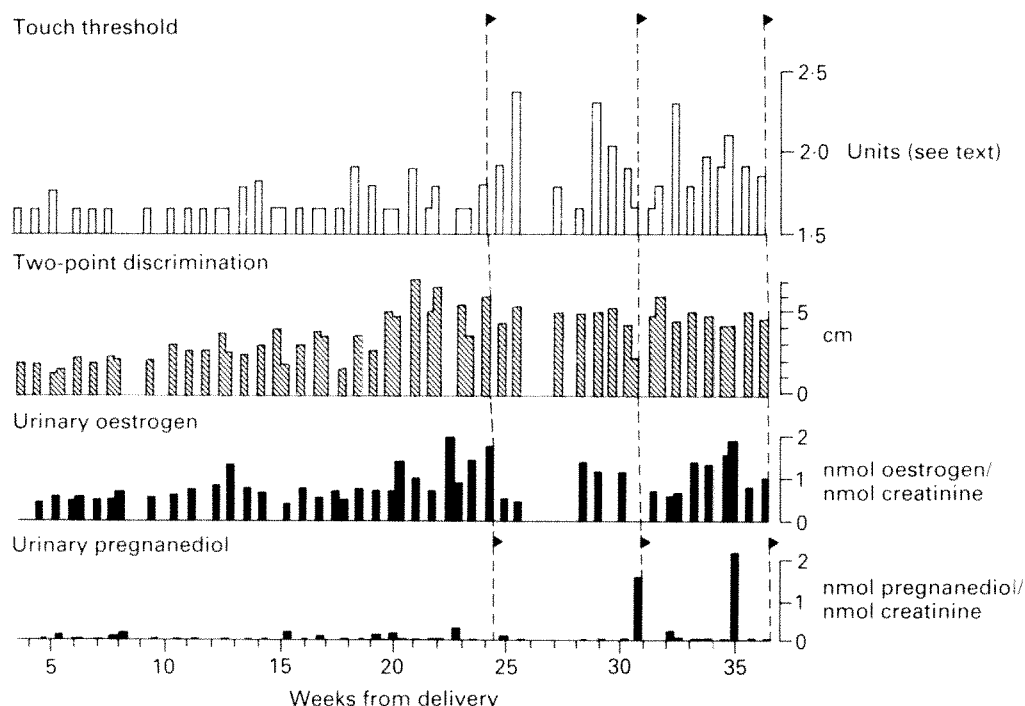


Fig. 1. Breast sensitivity measurements and urinary concentrations of ovarian steroids during the puerperium in one lactating woman. The vertical dashed lines with 'flags' indicate the onset of menstruation. Weaning was completed at week 35.

practical value in predicting the return of ovarian activity.

Acknowledgments

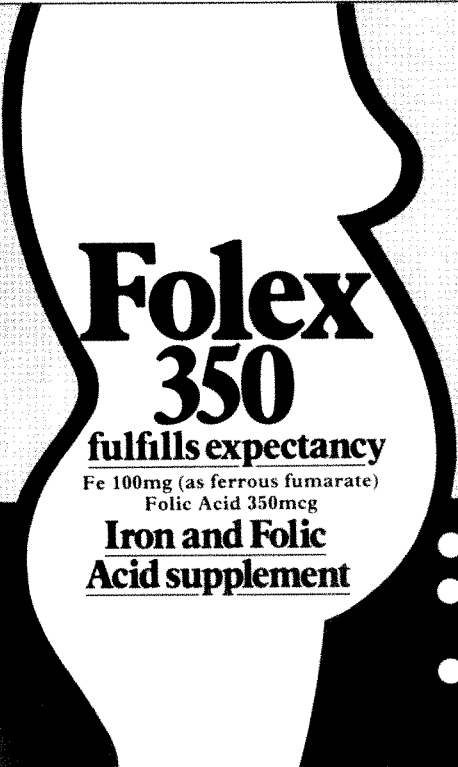
This investigation received financial support from the Special Programme of Research, Development, and Research Training in Human Reproduction, World Health Organization. Professor R. A. Hinde, FRS, kindly lent the aesthesiometer.

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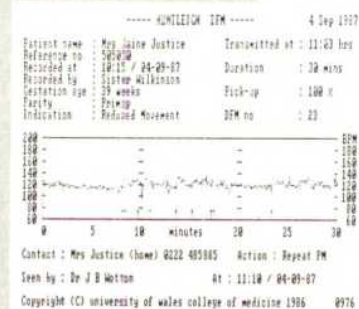
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CORRESPONDENCE

Early pregnancy termination: a comparison between vacuum aspiration and medical abortion using prostaglandin (16,16 dimethyl-trans- Δ_7 -PGE₁ methyl ester) or the antiprogesterone RU 486

Dear Sir,

We were very interested in the article by Dr Cameron and Professor Baird [*Br J Obstet Gynaecol* (1988) 95, 271-276].

We have been using Cervagem (gemeprost) pessaries to induce a medical abortion in women with missed abortion or a recognized fetal abnormality such as spina bifida or Down's syndrome diagnosed by either ultrasound scan, amniocentesis or chorionic villus sampling. The majority of pregnancies were mid-trimester.

Between 1986 and 1987, we treated 53 patients with Cervagem pessaries (gemeprost 1 mg) either for missed abortion in 21 or proven fetal abnormality in 32.

All the patients were admitted to a single ward adapted to their needs, providing private self-contained accommodation, with emotional and physical support during the in-patient period, and community support at home if required.

All patients were given one Cervagem pessary vaginally every 3 h until abortion took place, and there was no limit placed on the number of pessaries to be used.

In the missed-abortion group, the mean gestational age at diagnosis was 19 weeks (range 12-27 weeks) based on the last menstrual period. These patients required an average of three pessaries (range 1-5) before the fetus aborted. Of the 21 abortions 11 (52%) were complete and did not require evacuation of retained products, or blood transfusion; 10 (48%) required evacuation of retained products or manual removal of placenta under general anaesthesia, two of these patients required blood transfusion for excessive blood loss.

In the fetal abnormality group, where fetal death had not occurred, the same procedure was followed. In this group mean gestation was 18 weeks (range 12-23 weeks), the average number

of pessaries used was 4.4 (range 1-8). Of the 32 patients 18 (56%) aborted completely and required no transfusion or evacuation, in 14 (44%) the abortion was incomplete and required evacuation of uterus or manual removal of placenta, four of these patients required blood transfusion.

All the patients had adequate pain relief either with oral analgesics or intramuscular opiate. There were no major side-effects in any of the patients sufficient to cause the procedure to be abandoned.

Previous reports on uses of Cervagem in mid-trimester abortions have involved normal pregnancies (Nair *et al.* 1986; Sakamoto *et al.* 1982) and have reached favourable conclusions. Our experience in the use of Cervagem in pregnancies associated with fetal abnormality or early fetal death at a variety of gestational ages shows no major side-effects and avoids any operative procedures in just 50% of the patients studied.

We would therefore recommend the further assessment of this method of termination of abnormal and normal pregnancies at all gestations.

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the antiprogesterone RU 486 *Br J Obstet Gynaecol* 95, 271-276.

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A randomized trial of progestogens in the primary treatment of endometrial carcinoma

Dear Sir,

We were interested to read the report of the Yorkshire trial of adjuvant progestogen therapy in patients with recently diagnosed endometrial cancer [*Br J Obstet Gynaecol* (1988) 95, 166-174]. The number of patients accrued into the trial was chosen to give a 90% chance of detecting a 10% improvement at a 5% level of significance. Depending on the interpretation of '10% improvement', this means that for an expected survival of 75% in the control group, the authors were seeking a survival in the treatment group of either 82.5% or 85%.

We consider this to have been an unrealistic target.

Our experience with a higher risk endometrial cancer group is that more than 40% of deaths will be unrelated to the disease, a figure which is consistent with that quoted in the article (61% tumour deaths, 39% non-tumour deaths). If the expected death rate in controls is 25%, we can therefore assume that the expected death rate due to cancer is approximately 15% and that due to other causes is 10%. Assuming that the death rate due to other causes is the same in the treatment arm as in the control arm, then in order to reduce the overall death rate from 25% to either 17.5% or 15%, the cancer death rate needs to be reduced from 15% to either 7.5% or 5%. Even if there were no inoperable cases and all patients adhered to protocol this would be extremely optimistic.

A far more realistic target would be for a reduction in the death rate of about half of that

sought (i.e. from 25% to 12.5%). This is equivalent to a reduction in the cancer-specific death rate of 15% to 11%, and even for this much more modest expectation one would require approximately four times the number of patients entered into the Yorkshire trial.

The endpoint of the trial was survival rather than time to recurrence. It may be assumed that many of the control patients received progestogen therapy on recurrence and therefore the interpretation of the trial results could be that adjuvant therapy offered no survival advantage over therapy on relapse. We would be interested in the authors' comments on this proposition.

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Macdonald, R. R., Thorogood, J. & Mason, M. K. (1988) A randomized trial of progestogens in the primary treatment of endometrial carcinoma. *Br J Obstet Gynaecol* 95, 166-174.

Authors' reply

Dear Sir,

These points are well taken and we were probably over-optimistic in our expectations.

Treatment of recurrence was at the discretion of the local gynaecologist and many of the control patients did indeed receive progestogen therapy. Our recommendation was that progestogens should be reserved for recurrent lesions and it would be quite fair to restate that as adjuvant therapy it offers no survival advantage over therapy on relapse.

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BOOK REVIEWS

Baillière's Clinical Endocrinology and Metabolism International Practice and Research (Reproductive Endocrinology Vol.1 No.1) edited by H. G. Burger. Baillière Tindall, London, Philadelphia, 1987, 246 pages with figures and tables, £15.00 (UK & Eire), \$25.00 (USA & Canada), £18.50 (elsewhere)

The numbering of this book, No. 1 of Volume 1, suggests that it is the start of a new series from Baillière Tindall. If so, it augurs well, for the publishers have used the best formula for good multi-author scientific writing: pick someone who functions in the advancing front of research and leave him free to select authors who are themselves productive of new ideas and can write with skill and authority. Of course it is not everybody's cup of tea; it won't get anybody through the membership, and there is not much you could go out and try on patients tomorrow. It is a book for the researcher and the expert. Although the eleven chapters are on separate topics in reproductive endocrinology, they range about the central theme of the control of gonadal function. In view of the editor, it is not surprising that the book has a strong Australian flavour. It is all the better for that: Melbourne is one of the places where reproductive endocrinology is at.

In view of the uniformly high quality of the writing it is invidious to pick out particular reviews, but eleven chapters are too many to mention individually. Henry Burger did right to put the chapter on the pulsatility of reproductive hormones by Clarke and Cummins first. The demonstration of the pulsatile release of LHRH has created a new understanding of the hypothalamic-pituitary-gonadal axis. Who better to write about it than the people who did much of the original work.

I have hinted that the book is short on clinical application. It is retrieved from serious deficiency on this account by the chapters from Fraser and Baird on clinical applications of LHRH analogues and a short review of a new antiprogesterone, RU 486, by Garfield and Baulieu. The LHRH antagonists have proved

disappointing in the contraceptive field where much was hoped of them, but Fraser and Baird write enthusiastically about the agonists which are indeed finding increasing application in the suppression of gonadal function. It may not be too much to claim that we have at last in hand a safe means of temporary medical castration.

The progesterone inhibitor, RU 486, is the first of a series of antiprogestational steroids whose use in clinical practice is likely to extend well beyond their present use as abortifacients.

It is sometimes difficult to identify the level of knowledge of your readership. Garfield and Baulieu are the only authors in this book who may have underestimated their readership. In view of the level at which the rest of the book, and indeed the latter half of their own chapter, is written, I find it surprising that they should consider it necessary to include diagrams of the formula for progesterone and oestradiol and an undergraduate cartoon of hormone binding by receptor proteins in a target cell.

Our understanding of the paracrine and autocrine regulation of the gonads by factors generated within the gland is increasing rapidly. It is very difficult to write simply and with discrimination about an area where yesterday very little was known and tomorrow it may all change. Findlay and Risbridger manage to do that in an essay which I found the most scholarly in the book.

Arnold Kloppe

Clinical Obstetrics edited by Carl J. Pauerstein. John Wiley & Sons, Chichester/New York, 1987, 1017 pages with figures and tables, £38.45

This well-presented new American textbook is the work of more than 50 contributors under the editorship of Pauerstein. Most are from North America although there are two British contributions.

The book is intended to be a 'satisfactory guide to clinical practice', and is aimed particularly at house officers and physicians actively

engaged in obstetric practice. The chapters are grouped in order into sections on normal pregnancy and labour, antepartum and intrapartum fetal surveillance, problems specific to pregnancy and labour, maternal illness complicating pregnancy, and a final section on operative obstetrics.

Embryology is taken on without diagrams, but an excellent series of photomicrographs are well matched to the text. There is a comprehensible chapter on immunologic aspects of pregnancy. The chapter on antepartum care contains pictures of blank antenatal case sheets from hospital X—a form of presentation I always find unhelpful, however proud the authors are of their favoured system. A problem-oriented approach is advocated in which the pregnancy is Problem 1! This somewhat didactic chapter gives no thought to sharing antenatal care with other professionals, nor does it refer to the critical analyses of antenatal care from the Aberdeen workers.

The second section begins with a good practical review of fetal monitoring. The British eye has to adapt to the 3 cm/min paper speed in the examples. This is followed by a chapter on diagnostic ultrasound, and an excellent review of antenatal diagnosis of genetic disorders.

The third section opens with the editor's chapter on bleeding in pregnancy, which is thought-provoking, including a defence of home management in placenta praevia, and an admirably conservative approach to the management of placental abruption with a live fetus on admission. A whole chapter on premature rupture of the membranes may seem excessive but Gibb's review of this controversial subject is unlikely to be improved upon. A chapter on poor obstetrical outcome, which includes IUGR, congenital malformations, fetal wastage, macrosomia and non-immune hydrops is innovative and works well.

The chapter on breech presentation is detailed, well argued and well illustrated, and anticipates a decreased resort to caesarean section in the future. The next section on maternal problems in pregnancy is of generally high quality. There is a detailed and readable chapter on septic shock. The final section is on operative obstetrics. The chapter on episiotomy advocates the midline incision, and perhaps wisely includes diagrams of repair of third degree tear! The opinion is given that most primigravidae will be offered episiotomy, the principal indication

being 'to facilitate delivery'. The chapter on forceps and the vacuum extractor makes an interesting case for American obstetricians to overcome their reluctance to use the latter.

A concluding chapter refers to medico-legal consequences of obstetrics and includes the chilling information that in 1983 16% of all US obstetricians were involved in malpractice suits and in Florida 25% of obstetricians have abandoned the speciality! Overall, the book is a welcome addition to the list of postgraduate obstetric textbooks. The willingness of all the contributors to give credit to their predecessors by the widespread use of pertinent quotes from classical papers is most enjoyable. The membership candidate who is aware of the differences in transatlantic practice will find the book a useful adjunct to exam preparation, but I would recommend it most to the obstetrician in practice whose standard text on the office bookshelf has grown grey hairs.

Robert Fraser

Bacterial Vaginosis by W.I. van der Meijden. Van Gorcum, The Netherlands, 124 pages with figures and tables, Dfl 35

This short monograph written by a Dutch gynaecologist is a summary of a number of investigations relating to the prevalence, diagnosis and treatment of bacterial vaginosis (non-specific vaginitis). The opening chapter is an excellent résumé of current knowledge of this condition. The remaining chapters are in the format of scientific papers each addressing a specific topic. Each chapter has been published or submitted for publication elsewhere. The first two studies consider the problem of diagnosis, both clinical and microbiological. Dr van der Meijden uses different diagnostic criteria from other workers and this makes his data rather difficult to interpret; nevertheless, the material in these chapters is worthy of study.

The next chapter reports the results of a survey of women in general practices in which their experience of vaginal discharge was sought. This is followed by a laboratory study and three treatment trials. Unfortunately, different study designs, entry criteria and microbiological protocols prevent the results of each study treatment being compared directly.

It is a pity that the author did not restructure the text as the publication format has resulted in the repetition of information in the introduction

and methods section of each chapter. Furthermore, a single bibliography for the whole book would have been preferable to a list of references for each chapter.

This book will be of most interest to those directly involved in research on bacterial vaginosis, though the chapter on the general practice survey should be read by all those involved in the design of public health education programmes. At around £10.00 this book is a good buy for practising gynaecologists if only for the introduction, discussion and excellent photographs.

P. E. Munday

WHO Laboratory Manual for the Examination of the Human Semen and Semen-Cervical Mucus Interaction published on behalf of the World Health Organization by Cambridge University Press, 1987, 67 pages, £8.95

Skill in the examination and the analysis of semen is sadly lacking in many pathology laboratories. It was therefore a pleasure to see the publication of the second edition of the *WHO Laboratory Manual for the Examination of Human Semen and Semen-Cervical Mucus Interaction*, this time published by Cambridge University Press. This new edition comes in a sturdy plastic cover that should be sufficiently robust to stand up to prolonged use in a laboratory. The layout is similar to the previous edition but the print is slightly larger making it more attractive and also easier to read.

There have been some significant additions in this book. The number of colour illustrations has been increased and they are improved in quality. The section on sperm-cervical mucus interactions has been considerably extended and includes some excellent illustrations. The appendix has also been enlarged. It contains the methodological details of many useful staining techniques. Included also are the methods used for the detection of antisperm antibodies, and the mixed antiglobulin test and the immunobead test are both described in full.

Too often the analysis of semen is limited to the examination of the sperm only and scant attention is paid to the other aspects of seminal fluid. With the increasing awareness of the need to examine for abnormalities in the secretions of the accessory glands, it was of interest to see the inclusion of methods for the measurement of zinc, fructose and citric acid in seminal fluid in

this edition of this book. A method for the determination of ATP in semen is also to be found in the appendix of this edition of the manual. Although the estimation of ATP is only rarely undertaken in many andrology laboratories, such an estimation can indeed be useful clinically. It is therefore hoped that such analyses will, in time, become increasingly common. Included also for the first time in this edition, are the commonly used methods for the culture of bacteria in semen which will be useful information for any laboratory that carries out semen analyses.

The protocol for the zona-free hamster oocyte test is described in full in this edition of the WHO manual. Although such techniques will only be available to those laboratories with access to an animal house, it is good to see these tests of sperm function now being included in standard books on the analysis of semen. This manual concludes with a bibliography that contains a number of important review articles of semen.

This little book is excellent value for money and should be included in the libraries of all laboratories involved in the examination of semen. It has helped a great deal in the past to improve the standard of semen analysis and this new edition continues this tradition.

Anne M. Jequier

The Contraceptive Ethos (Philosophy and Medicine 27) edited by Stuart F. Spicker, Williams B. Bondeson & H. Tristram Engelhardt Jr. D Reidel Publishing Co., Dordrecht, 254 pages, £32.95

This volume, subtitled *Reproductive Rights and Responsibilities*, developed from a symposium of the same name which took place in 1983. The long gestation was unfortunate since contraceptive freedom was severely affected by the AIDS epidemic in the intervening years, a fact acknowledged in the introduction.

The four sections of the book deal with contraception in terms of history, rights, responsibilities and sexuality. The new contraceptive ethos is not defined: the whole book leaves an impression of its subject and the conclusions reached are all rather nebulous and negative. This is, after all, a philosophical work rather than a medical text. World population growth and population control are discussed, but the

authors are not agreed on the need for an international contraceptive ethos. The final chapters conclude that contraception is not a means of controlling personal destiny and that it plays virtually no role in redefining sexuality.

The authors, and the resulting book, are unashamedly American. One chapter concerns the average American family planning clinic nurse, a 42-year-old Protestant who disapproves of premarital sex. Another, entitled 'The Right to Fornication', is concerned entirely with American case law. Chapter titles like 'A Womb of One's Own' are jokey and so is some of the content: Solomon reminds us that sex is not just matter in motion, sex is ideas. However, every chapter is carefully argued and well written, a rare feat in a multi-author work.

Those interested in family planning will find this excursion into the philosophy of sex and contraception interesting but not very informative. The book gives a narrow and foreign view of a worldwide subject and its value, other than as a curiosity, is not great. As Spicker says in the introduction: sex on satin surpasses sex on paper.

M. I. Archer

Books received some of which may be reviewed at a later date

Urinary Tract Infection and Laboratory Practice by Rosalind Maskell. Edward Arnold, London, 1988, 268 pages with figures and tables, £25.00

Sectional Fetal Anatomy in Ultrasound by A. Staudach. Springer Verlag, Berlin, Heidelberg, New York, 1987, 198 pages, 247 figures, DM 120

Perinatal Events and Brain Damage in Surviving Children edited by F. Kubli, N. Patel, W. Schmidt & O. Linderkamp. Springer Verlag, Berlin, Heidelberg, New York, 1988, 338 pages, 51 figures, 173 tables, DM 120

Fetal Echocardiography: An Atlas by Kathryn L.

Reed, Caroline F. Anderson & Lewis Shenker. Alan R. Liss, Inc., New York, 1988, 146 pages fully illustrated, \$62.50

Drug Use in Pregnancy Second edition by Jennifer R. Niebyl. Lea & Febiger, Philadelphia, 1988, 245 pages with figures and tables, \$29.00

Treatment and Prognosis Obstetrics and Gynaecology edited by J. G. Grudzinskas & T. Beedham. Heinemann Medical Books Ltd, London, 1988, 257 pages, £17.50

Cancer Genetics in Women Volumes 1 and 11 edited by Henry T. Lynch & Stig Kullander. CRC Press, Inc., Florida, 1987, both volumes illustrated with figures and tables, £146.50

Textbook of Uncommon Cancer edited by C. J. Williams, J. C. Krikorian, M. R. Green & D. Raghavan. John Wiley & Sons, Chichester, 1988, 1064 pages with figures and tables, £110.00

Contemporary Issues in Fetal and Neonatal Medicine. 3. Ethical Issues at the Outset of Life edited by William B. Weil Jr & Martin Benjamin. Blackwell Scientific Publications, Oxford, 1987, 271 pages, £36.00

Essential Reproduction Third edition by Martin Johnson & Barry Everitt. Blackwell Scientific Publications, Oxford, 1988, 377 pages with figures and tables, £14.50

A Pocket Obstetrics and Gynaecology Eleventh edition by Stanley G. Clayton & John R. Newton. Churchill Livingstone, Edinburgh, 1988, 382 pages with figures and tables, £8.95

Human Behaviour in Illness Fourth edition by Lynn Gillis. Faber & Faber Ltd, London, 1988, 216 pages, £4.95

Principal Drugs Ninth edition by S. J. Hopkins. Faber & Faber Ltd, London, 1988, 248 pages, £2.95

Having A Baby by Nancy Kohner. BBC Books, London, 1988, 232 pages illustrated, £10.95

COMMENTARY

Dysmenorrhoea

This issue publishes a report by Hauksson *et al.* (p. 898) on the effects of vasopressin and a synthetic antagonist on myometrial activity and uterine blood flow. These workers have previously investigated many aspects of the influence of vasopressin on the uterus. The antagonist, an analogue of oxytocin, counteracts the increased myometrial activity and vasoconstriction provoked by vasopressin and the authors conclude that this antagonist could be used to treat primary dysmenorrhoea.

Dysmenorrhoea is an important clinical problem which affects about 50% of women. Primary dysmenorrhoea occurs in the absence of pelvic pathology while secondary dysmenorrhoea is usually associated with pathology such as pelvic inflammatory disease, endometriosis or submucous fibroids. Women with primary dysmenorrhoea used to be considered neurotic until it became clear that the condition was associated with uterine hypercontractility leading to uterine ischaemia (Lundstrom 1981).

Many aetiological factors have been implicated in the hypercontractility of primary dysmenorrhoea. Pickles *et al.* (1965) showed that prostaglandins (PG) were involved, they found elevated concentrations of PGE₂ and PGF_{2α} in endometrium and menstrual fluid and noted higher levels and a raised PGF_{2α}/PGE₂ ratio in women with primary dysmenorrhoea than in pain-free controls. Since the original work by Pickles, many investigators have found dysmenorrhoea to be associated with elevated PGF_{2α} levels in menstrual fluid, endometrium and endometrial jet washings (Rees 1988). However, a correlation between PGF_{2α} concentrations in menstrual fluid and uterine contractility has only been found on day 2 of menstruation (Lumsden *et al.* 1983). Leukotrienes, compounds allied to prostaglandins, have recently been implicated in dysmenorrhoea, with elevated levels of leukotrienes C₄,

D₄ and E₄ being found in endometrium, but not myometrium, of women with dysmenorrhoea (Rees *et al.* 1987). During menstruation, plasma vasopressin levels are higher in dysmenorrhoeic women than in controls (Åkerlund *et al.* 1979). Vasopressin infusion stimulates uterine activity, decreases uterine blood flow and causes dysmenorrhoea-like pain and, *in vitro*, it constricts uterine arteries (Åkerlund *et al.* 1983). Vasopressin may therefore be involved in the aetiology of dysmenorrhoea.

Interaction between vasopressin, prostaglandins and leukotrienes has been suggested. Vasopressin infusion has been found to produce an increase in PGF_{2α} metabolite levels in peripheral venous plasma (Strömberg *et al.* 1983). These metabolites were thought to come from the uterus, but the effect of vasopressin on menstrual fluid PG levels was not examined. Plasma vasopressin levels remain elevated in women whose dysmenorrhoea is successfully controlled by PG synthase inhibitors (Strömberg *et al.* 1981), suggesting that vasopressin acts by stimulating PG synthesis. On the other hand, the same group found that the stimulatory effect of vasopressin infusion on uterine activity was similar in the presence or absence of a PG synthase inhibitor, although PGF_{2α} metabolite levels were reduced by the inhibitor (Strömberg *et al.* 1983). These findings suggest that although vasopressin may stimulate PG production, it must also have a direct effect on the myometrium. A possible interaction with leukotrienes is suggested by recent studies in which contractions induced by vasopressin in rat myometrium were inhibited by a chemical inhibitor of lipoxygenase activity, nordihydroguaretic acid (Demers *et al.* 1984).

Although excessive levels of prostaglandins, leukotrienes and vasopressin have been found in primary dysmenorrhoea, the primary stimulus for their production remains unknown.

A wide range of treatments has been proposed for primary dysmenorrhoea over many years and present medical treatment consists mainly of PG synthase inhibitors or the combined oestrogen-progestogen contraceptive pill. PG synthase inhibitors in current use include mefenamic acid, naproxen, ibuprofen and indomethacin. Most studies have shown them to be effective in reducing menstrual pain in 80–90% of patients (Dawood 1984). During treatment, intrauterine pressure recordings show a decrease in uterine contractions. The combined oral contraceptive pill is also effective in 80–90% of women (Kremser & Mitchell 1971), and probably acts by reducing the capacity of the endometrium to produce prostaglandins (Chan *et al.* 1981).

Concern remains about the 10–20% of the patients with primary dysmenorrhoea who fail to respond either to PG synthase inhibitors or to oral contraceptives. Some of these women may really be suffering from secondary dysmenorrhoea with pelvic pathology, requiring appropriate investigation, but the concern has led to examination of new agents. Dr Åkerlund's group has explored many vasopressin antagonists in recent years (Åkerlund *et al.* 1986; Melin *et al.* 1986). The most effective so far appears to be 1-deamino-2-D-Tyr (OEt) 4-Thr-8-Orn-oxytocin, which is considered in the article in this issue (p. 898). In-vivo administration by bolus injection of the antagonist inhibits vasopressin-induced reduction in endometrial blood flow, stimulation of uterine activity and dysmenorrhoea-like pain. Endometrial blood flow was estimated by a thermistor technique and myometrial activity by intrauterine pressure recording. In-vitro studies on uterine arteries show that the antagonist also inhibits vasopressin-induced vasoconstriction. Åkerlund (1987) reported that in a placebo-controlled trial the antagonist alleviated dysmenorrhoea in 50% of the women.

Currently available vasopressin antagonists are not suitable for the clinical treatment of primary dysmenorrhoea. Studies have only been of a single intravenous injection with a short-lasting effect, since the half-life is about 17 min (Lundin *et al.* 1986). Intranasal administration of posterior pituitary hormones is common, but the bioavailability of 1-deamino-2-D-Tyr (OEt) 4-Thr-8-Orn-oxytocin in its present form is poor (Åkerlund *et al.* 1986).

Further studies are required using different doses, routes of administration and delivery

systems before vasopressin antagonists can be recommended for clinical use in dysmenorrhoea. Even then they may prove only of limited value since they seem to be less effective than PG synthase inhibitors or oral contraceptives. Perhaps they may be helpful in women unresponsive to the other treatments. If so, they will be a very useful addition to the treatment of dysmenorrhoea.

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Antenatal care; current practice in debate

MARTIJN P. HERINGA, HENDRIK J. HUISJES

Summary. The antenatal screening practice of 67 teaching hospitals in the European Community was assessed by use of a written questionnaire. There were minor differences in mean numbers of tests performed in different countries, but only limited agreement was found on the selection of tests that were performed. There was no unity of practice within the countries. No correlations were found between the number of screening tests performed and the size of the units, or the composition of the attending staff.

The aim of antenatal care is to reduce perinatal mortality and morbidity and also maternal complications associated with pregnancy and childbirth. To allow early intervention, identification of risk factors for adverse outcome of pregnancy is mandatory (Cuckle & Wald 1984). Although medical and obstetric history will primarily direct the antenatal examinations, most pregnant women are subjected to a number of tests as routine screening procedures.

The variation across and within countries in the number and range of such tests was the subject of a substudy of the larger European Community (EC) Collaborative Study on Pre-, Peri- and Postnatal Care Delivery Systems.

Subjects and methods

In 1985 an enquiry was conducted among university and major training hospitals in the EC. In each country, a representative selected the units, if possible evenly spread across the country. The heads of the obstetric departments were requested to co-operate and to fill out the enquiry form.

The enquiry contained 27 questions about antenatal examination practice. The subjects and examinations which were assessed are listed in

Table 1. The items are clearly very heterogeneous, as they concern various maternal and fetal risk factors. For the sake of brevity all items will be referred to as 'tests'.

The clinicians were asked whether each of those tests was either routinely performed, or not performed at all, irrespective of the obstetric risk as assessed from history or physical examination. If neither alternative fitted—for instance, routine application was the unit's official policy, but the test was often omitted in practice—'recommended' could be a third possible answer.

If the test was routinely performed, additional information was obtained about the technique used, the frequency of the test and the gestational age at which it was applied.

General data were collected about the average numbers of bookings, antenatal visits and deliveries, and on the professional qualifications of those providing the antenatal care in the units.

Verification of the obtained data was not possible due to practical problems.

Results

In 1985 the EC consisted of 10 member states. For practical reasons Belgium and Luxembourg were taken together. Though the aim was to enrol 10 units from each country into the study, the total number was limited to 67. Of those, 46 units were university hospitals.

In some countries fewer than 10 units fitted the selection criteria; in others the willingness to co-operate was less than expected. No further

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information was available about the units that were asked and did not take part in the study. An exact response rate can therefore not be calculated.

Size and number of the units in each country are shown in Table 2.

The mean number of bookings was 2740 per unit (range 300–8150). All units together provided the antenatal care for over 160 000 women per year. There was a remarkable variation in the number of visits (2–15), even between units in the same country.

Further analysis showed no relation between the number of visits and the number of overall bookings for antenatal care, nor a consistent difference in the number of antenatal visits between university and other hospitals.

The mean scores of the countries on all tests are presented in Table 3. Fewer than 2% of the questions were left unanswered. Of the 30 tests 14% were scored as 'recommended', but this appeared to have different meanings for each test and therefore no general conclusions could be drawn from these answers. The remaining 83% of the answers clearly indicated either 'routine' or 'not routine' performance of the tests. The mean

number of tests routinely performed in all units was 17 (SD 3), with the highest mean scores in Italy and Belgium, the lowest in Denmark and Ireland. The standard deviation of some of the scores was relatively high. This indicated a considerable range of values within the countries. In almost all cases the median value was equal to the mean score.

Further analysis aimed to find out if there were units with a more restrictive or a more active testing policy. The distribution of units divided in groups by the number of tests routinely performed is presented in Fig. 1. The shape of the histogram indicates that some 10% of the units performed 75% or more of the tests as a routine procedure. With one exception all were university hospitals in Belgium, Italy, West Germany and the UK.

Table 4 presents the policy of the majority of all units for each test. There appeared to be general agreement on the routine performance of the first 11 mentioned tests and subjects. In this way, however, a comparison of the practice in different countries was not possible. Therefore for every test and for each country the proportion of units that did or did not carry out the test as a routine

Table 1. All 30 'tests' (subjects and disorders) listed in the questionnaire

Breast examination	Coagulation disorders	Placental function tests
Vaginal examination	Blood pressure	Maternal α -fetoprotein
Cervical smear	Weight	Gestational diabetes mellitus
Asymptomatic bacteriuria	Proteinuria	Syphilis
Blood group/rhesus (father)	Glycosuria	Toxoplasmosis
Blood group/rhesus (mother)	Microscopic urinalysis	Group B β -haemolytic streptococci
Haemoglobin	Fundal height	<i>Listeria monocytogenes</i>
Haematocrit	Fetal movements	Rubella
Ferritin	Ultrasound	Hepatitis B-antigen
Haemoglobinopathies	Cardiotocography	Cytomegalovirus

Table 2. Inventory of co-operating countries and extent of antenatal care

Country (abbreviation)	No. of units	Mean no. of bookings (range)	Mean no. of visits (range)
Belgium, Luxembourg (B + L)	7	1240 (750–2000)	9 (8–12)
West Germany (D)	10	1500 (700–2000)	9 (5–12)
Denmark (DK)	7	2640 (1750–3400)	5 (2– 8)
France (F)	8	2370 (1100–3000)	6 (4– 8)
Greece (GR)	4	3310 (1500–7000)	6 (3–10)
Italy (I)	5	2370 (850–2900)	7 (5– 8)
Rep. of Ireland (IRL)	7	4850 (2000–8150)	8 (4–10)
The Netherlands (NL)	9	920 (300–1500)	11 (8–15)
United Kingdom (UK)	10	4120 (2500–5600)	6 (4–10)
Total (range)	67	2740 (300–8150)	7.6 (2–15)

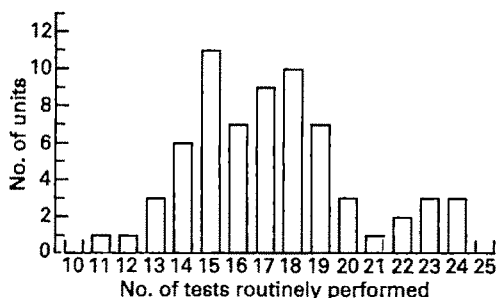


Fig. 1. Distribution of units divided into groups by the number of tests that were routinely performed ($n = 67$).

procedure was calculated. Next, for each test the mean and standard deviation on these proportions was computed, and the coefficient of variation (CV) could be derived. The CV of this mean reflects the range of the proportions and can be used as a measure of agreement on practice in different countries (Table 4).

When the CV is also taken into account, relatively high values ($>20\%$) were found even for 5 of those 11 tests for which at first sight a high degree of agreement was found. This means that there were considerable differences in practice between countries, in spite of the apparent similarity. This was even more pronounced with most of the other examinations where much higher CV values were found.

Although additional data were obtained on the frequency with which tests were performed and on the techniques used, those results were presented elsewhere (Heringa & Huisjes 1988).

Finally, the results of the enquiry were examined in relation to the attending staff in the hospitals. Residents took part in antenatal care in 65% of the units and midwives in 53%. There were no important differences in mean number of visits, number of routinely performed tests or number of deliveries in relation to composition of the attending staff.

Discussion

As official regulations on antenatal care do not always reflect what is done in practice, the choice was made to assess the actual policies of university and major training hospitals. The level of obstetric care offered in those hospitals was taken as the best indication of what was representative for the countries; but was it?

From the data presented on the extent of the obstetric care it appeared that mainly larger units

were involved in the study. Such centres usually also provide specialized obstetric care for a selection of women with high-risk pregnancies. The distribution of risk factors in the populations of those hospitals is therefore often different from the average in the country. Besides, this may result in a more active attitude towards screening even where low-risk pregnancies are concerned. In addition, scientific interest may have an influence on screening practice. Indeed, in this study the most active screening practice was found in some of the university hospitals.

All these factors might lead to an impression of the antenatal screening practice which is not fully representative for the whole country. Moreover, the comparability of the mean results per country may be limited because of differences between health care systems.

In an attempt to reduce this selection bias, the clinicians were explicitly asked to report on the screening practice as it was routinely performed irrespective of the individual obstetric risk. Furthermore, the results showed that the mean number of visits was about the same as reported by others (WHO Perinatal Study Group 1985) for the whole of Europe. Although the range of values of the individual countries indicated that there were important differences within as well as between countries, this concurs with the results of a previous study (Blondel 1986). Therefore, in this respect the group of units co-operating in this study is not different.

Finally, as primarily teaching hospitals were selected—where future midwives, doctors and obstetricians are trained—the results of the enquiry may at least give a fair impression of practice considered advisable in antenatal care in the EC.

The study considered only those antenatal screening tests which were routinely performed irrespective of individual obstetric risk. With only two clinicians mentioning additional examinations, the list of tests in the questionnaire seemed virtually complete. It may be reasonable to assume that there were no important differences in diagnostic facilities between and within the countries, since all were teaching hospitals. Also, taking into account that the mean number of tests routinely applied was almost the same in all countries, it is remarkable that only limited agreement was found on the selection of examinations actually performed. Apparently optimal results in obstetric care were thought to be achieved with very different policies of antenatal

Table 3. Mean score (and SD) of each country on the number of tests performed or not

Country	Test not routine	Test as routine	Test recommended	Answers missing
B + L	5.3 (2.3)	20.1 (2.9)	3.9 (3.8)	0.7 (1.0)
D	6.9 (4.4)	18.1 (2.6)	4.1 (4.1)	0.9 (1.4)
DK	11.0 (4.5)	14.6 (2.6)	3.5 (3.9)	0.8 (1.2)
F	9.0 (3.4)	16.0 (1.9)	4.3 (3.2)	0.5 (1.1)
GR	7.8 (5.3)	16.5 (1.7)	5.5 (3.6)	0.3 (0.5)
I	2.4 (2.6)	21.0 (3.9)	5.8 (1.6)	0.8 (1.3)
IRL	10.7 (3.1)	15.4 (2.2)	3.0 (1.2)	0.7 (0.8)
NL	9.9 (2.9)	15.8 (2.0)	3.9 (4.0)	0.4 (0.7)
UK	7.8 (3.7)	18.0 (2.1)	3.9 (3.7)	0.3 (0.7)
Total mean (SD)	7.9 (2.8)	17.0 (3.0)	4.1 (0.9)	0.6 (0.2)
(% of all 30)	(26)	(57)	(14)	(2)

Table 4. List of all tests, divided in columns and rank order of the majority policy of all units

Routine in over 50% of the units		Not routine in over 50% of the units		No majority
Blood pressure	(100/0)	<i>Listeria monocytogenes</i>	(75/37)	Hepatitis B-antigen
Glycosuria	(100/0)	Ferritin	(69/31)	Cardiotocography
Weight	(98/3)	Cytomegalovirus	(62/59)	Maternal serum
Blood group/rhesus (mother)	(98/4)	Group-B haemolytic streptococci	(61/42)	α -fetoprotein
Fundal height	(97/8)	Placental function tests	(57/38)	Microscopic urinalysis
Syphilis	(97/27)	Blood group/rhesus (father)	(55/69)	
Proteinuria	(97/12)	Coagulation disorders	(54/35)	
Haemoglobin	(90/21)	Haemoglobinopathies	(54/68)	
Vaginal examination	(88/21)	Toxoplasmosis	(52/80)	
Ultrasound	(82/22)			
Rubella	(82/52)			
Gestational diabetes mellitus	(66/35)			
Fetal movements	(64/26)			
Cervical smear	(63/51)			
Haematocrit	(59/69)			
Asymptomatic bacteriuria	(54/76)			
Breast examination	(51/43)			

The figures in parenthesis show the percentages of all 67 units/coefficient of variation (CV) of the mean (x) of the proportions of the units in each country performing tests routinely or not ($CV = \frac{SD}{x} \times 100\%$).

care. It was as if screening activity was seen to be beneficial, regardless of which tests were used.

Agreement over routine screening was restricted to the more traditional tests. Disagreement mainly concerned the modern techniques, with the exception of ultrasound. The divergence among screening policies might therefore be interpreted as a sign of uncertainty about the value of the tests. Doubts about the benefits of a number of antenatal screening tests are understandable (Grant & Mohide 1982). On the one

hand, the effectiveness of early treatment is limited in the most frequent disorders of pregnancy; on the other hand, the incidence of disorders that can be treated effectively at an early stage is low. Both factors result in only minor effects on outcome at the population level. Besides, screening may have disadvantages, for example the anxiety caused by false-positive results, where sufficient reassurance cannot always be offered.

Furthermore, although the value of some antenatal screening tests may be obvious in individual

cases, the efficacy of only a few can be (and has been) assessed for the population (Enkin & Chalmers 1982).

Contrasting the widely divergent policies with the small differences in outcome, even in adjacent countries (Blondel *et al.* 1985), it is clear that more research on the epidemiology of pregnancy disorders and on antenatal screening is essential. The need is even more compelling because the financial resources for antenatal screening may be increasingly limited in almost all EC countries (Muir Gray 1984).

Conclusion

As far as the results are representative of each country, it must be concluded that there is little agreement on practice in antenatal screening in the EC. Factors explicitly associated with differences in practice were not found in the available data. There are important differences in antenatal health care delivery systems in the EC countries (Blondel *et al.* 1985) and, of course, the effects of such differences cannot be discounted. But in this study they were not reflected in significant differences in intensity of antenatal care. This may be due to the fact that the practice of the units was often widely divergent, even within countries.

The finding that on average as many as 55% of the current screening tests are performed as a routine procedure irrespective of the obstetric risk, with only slight overlap of tests, indicates the uncertainty of the clinicians about the value of antenatal screening. The benefits of routine screening, especially in a low-risk population, are probably overestimated and the disadvantages undervalued. In addition, the presumed benefits of screening might even result from other associated factors, rather than from the tests performed. Further research on the effects of antenatal screening programmes is necessary to improve a situation which involves so many women all over Europe. As large samples will be needed, international collaboration will be required to establish results within a reasonable time. Standardization of diagnostic procedures and of the registration of epidemiological data

should form the basis needed for this co-operation. It is for the EC to take initiating and co-ordinating action in this field.

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Perinatal data reliability in a large teaching obstetric unit

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Summary. In this inter-rater agreement study of antenatal and neonatal variables collected in a large teaching obstetric unit, information routinely collected by hospital staff was compared with that collected by a specially trained physician and a social worker. Agreement between the two sources of data was evaluated using kappa statistics and intraclass correlation coefficients. Excellent agreement was observed for some variables such as maternal and newborn anthropometric measures, and previous birthweight, but there was poor agreement for others such as indicators of physical activity, work during pregnancy and blood pressure measures. Some of the limitations are due to problems in phrasing questions, patients' recall, interviewer bias and abstracting data. We recommend that epidemiological studies should always include a reliability component, proper standardization of personnel and instruments and include, when published, validity data and examples of questions used.

Data routinely collected in obstetric services have been the basis for reports describing several aspects of antenatal care in general (Chalmers *et al.* 1976b; Rahbar 1985; Faundes *et al.* 1982), its assessment (Ryan *et al.* 1980; Sokol *et al.* 1980), the development of antenatal risk scores (Graziano *et al.* 1981) or for medical audit

(Chng *et al.* 1980). This source of information is useful because many hospitals and health regions now have precoded and computerized medical records which provide a relatively simple, rapid and inexpensive method for monitoring and assessing perinatal services and exploring epidemiological questions. Furthermore, the aggregation of several of these datasets permits the study of less frequent pathologies that would otherwise require long periods of data collection.

Obstetric data are collected by numerous personnel, including doctors, midwives, nurses, social workers and clerks. Standardized data collection procedures are frequently not used in such a context, even within epidemiological research projects and the quality, reliability, and accuracy of the information have rarely been investigated for perinatal data. Attempts in this direction have been limited to very specific issues (Little *et al.* 1984) or techniques (Yobs *et al.* 1985; Wilcox & Horney 1984; Peck 1980; Halperin *et al.* 1985; MacGillivray *et al.* 1969), or

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to preliminary assessments (Cherkin *et al.* 1984). The experience of other medical disciplines has not been followed (Herrmann 1985; Koran 1975).

We present here a study of inter-rater agreement performed to assess the reliability of data collection in a large teaching obstetric unit in Guatemala City, Guatemala.

Subjects and methods

The study population was selected from the Guatemalan Social Security Institute's Gynaecology and Obstetrics Hospital. This is a 230-bed hospital with a neonatal intensive care unit, staffed by residents, middle level attending physicians and consultants in obstetrics and gynaecology. The women delivered in this hospital are those eligible for social security benefits, women formally employed and wives of employed men.

Between 1 April 1984 and 10 January 1986 all pregnant women who attended the hospital for their first antenatal visit were enrolled in the Guatemalan Perinatal Follow-up Study ($n = 16\ 850$). This is a prospective follow-up study of risk factors associated with negative pregnancy outcome in this lower middle class urban population in Guatemala City. Women with no antenatal care at all and those having antenatal care at other peripheral clinics were excluded. The organization, population and methodology of this study have been described in detail elsewhere (Kestler *et al.* 1987). The data collection instrument used in the study is available upon request.

During the first 6 months of the perinatal follow-up study, the field director met with clinic personnel every 2 weeks to standardize measuring procedures. The objectives of these procedures were to minimize undesirable sources of variation and to enable evaluation of the magnitude of error in estimates. The field director and a physician or nurse each measured 10 pregnant women twice during the same antenatal visit. Analysis of variance for repeated measures was performed and the standardization procedure was considered complete when all sources of variation other than the subject were statistically non-significant.

The inter-rater agreement study we present here was conducted after the first 6 months of standardization. Participants were selected from the population of women enrolled in the peri-

natal follow-up study. At a non-predetermined time each day during the study period, the project resident asked the clinic clerk, who was unaware of the study objectives, to select from each of the eight antenatal clinics the next patient completing a visit. This patient was then taken to the study clinic for a repeat antenatal visit and interview. The project resident also attended the labour and delivery suite during selected mornings. During that period, any woman in labour was followed up by him until she was delivered. Data were simultaneously collected by the project resident and the attendant physician during labour; at birth the project resident independently determined the Apgar score and repeated all the anthropometric measurements for the infant.

The study thus comprises four sections defined by the type of variable investigated: socio-demographic, obstetric history, antenatal care, and labour and delivery. Because samples of women for the different sections of the study were selected separately, women who participated in one part of the inter-rater agreement study did not necessarily participate in the other parts.

Information routinely collected for the perinatal follow-up study by hospital staff was used as one source of data for the assessment of inter-rater agreement. A physician and a social worker on the research project were the other source of data. Results from one rater were not available to another. It was unclear whether the physician and social worker employed by the inter-rater agreement study ascertained and recorded information more accurately than hospital staff who had also been trained in standardized data collection procedures for the follow-up study. Therefore, analyses of inter-rater agreement were performed considering all raters equally qualified.

Analysis of inter-rater agreement for categorical variables was based on calculation of proportion of agreement and kappa. Kappa is a measure of agreement corrected for chance and is calculated as $(Po - Pe)/(1 - Pe)$, where Po is the observed proportion of agreement and Pe is the proportion of agreement expected by chance. Perfect agreement is indicated by kappa equal to one and chance agreement is indicated by kappa equal to zero. Positive values of kappa indicate better than chance agreement and negative values indicate less than chance agreement. Guidelines have been published with the follow-

ing suggested interpretation for kappa: >0.75 represents excellent agreement beyond chance, $0.40-0.75$ represents fair to good agreement beyond chance, and <0.40 represents poor agreement beyond chance (Fleiss 1981).

For continuous data, the intraclass correlation coefficient was used as a measure of inter-rater agreement. Except for a factor involving $1/n$ (n = number of subjects), the intraclass correlation coefficient is equivalent to a weighted kappa and can be interpreted similarly to it (Fleiss 1981).

Results

Table 1 presents descriptive statistics for participants of the inter-rater agreement and follow-up studies. Women who participated in the inter-rater agreement study had a mean age of 26 years and completed a mean of 6.1 years of school. They were similar to the population of women who participated in the follow-up study on all variables except infant birthweight ($P < 0.001$) and the presence of potable water in the house ($P < 0.05$).

Table 2 presents data on agreement between hospital medical staff and social workers (only for socio-demographic variables) and the research project physician and interviewer (for socio-demographic variables and obstetric history). Education, income and an indicator of crowding in the household (persons/room) had good to excellent agreement. Agreement was also high for husband's occupation. However, the value of kappa was small, indicating that much of the observed agreement for this variable could have been due to chance. Agreement on house characteristics was excellent for the presence of water and sanitation but only fair for the type of floor and the presence of a television set.

History of obstetric complications, birthweight of the previous infant and birth interval showed excellent agreement, but gestational age of the previous infant showed poor agreement.

Antenatal data were ascertained during the 29th week gestation. Data on agreement for antenatal variables collected by physical examination and interview are presented in Table 3. Anthropometric measurements, including height, weight and uterine height, showed excellent agreement. However, blood pressure measurements exhibited poor agreement. Identification of the presenting part and presence of oedema and infection showed high proportions

of agreement. However, kappas indicated poor agreement beyond chance. Determinations of gestational age and pre-pregnancy weight by interviews showed excellent agreement. Other antenatal interview data also showed very high proportions of agreement. With the exception of smoking, which could be explained almost totally by chance, agreement beyond chance for these variables was fair to good.

Labour and delivery and newborn characteristics are presented in Table 4. The proportion of agreement was high for all labour and delivery variables. For use of anaesthesia and type of delivery, values of kappa indicate that most of this agreement could not be explained by chance. However, for use of oxytocin practically all the observed agreements could have been due to chance. Birthweight, length and head circumference showed excellent agreement. Gestational age, estimated either from last menstrual period or by physical examination using a shorter version of the Dubowitz assessment, also exhibited excellent agreement. Fair to good agreement was observed for the Apgar score at 1 min and excellent for the 5-min Apgar score.

Data collected on physical activity are presented in Table 5. Overall, type of activity had a high proportion of agreement and a high kappa, indicating that the observed agreement could not be explained by chance. Information on physical activity in the home also had high proportions of agreement. Values of kappa indicated fair to good agreement beyond chance. Finally, information on whether the woman held a paid job exhibited excellent agreement. However, variables that assessed physical activity at work had poor to fair agreement (Table 5).

Discussion

We have compared information from two sources: (i) hospital records completed by physicians, nurses, and social workers which were abstracted immediately after each antenatal visit and delivery to a precoded data collection form used in the follow-up study; and (ii) information collected by a trained resident obstetrician and a social worker participating in the inter-rater agreement study. Thus, reliability refers to the data recorded on the data collection form for the follow-up study. It is assumed, and will not be discussed here, that the abstracting and coding processes were without error or at least that the error was randomly distributed. Nevertheless,

Table 1. Selected descriptive statistics of the inter-rater agreement and follow-up studies

Variable	n	Inter-rater agreement†			Follow-up study‡		
		Mean	(SD)	%	Mean	(SD)	%
Maternal age (years)	140	26.0	(5.5)		25.3	(5.5)	
Years schooling	140	6.1	(4.7)		5.9	(3.9)	
Unmarried (%)	140			2.1			3.8
No. of persons in the house	140	4.5	(2.7)		4.1	(2.2)	
No. of persons per room	140	3.1	(1.4)		3.1	(1.4)	
No potable water in house (%)	140			12.9			19.8*
No sanitation (%)	140			2.1			2.3
Working mother (%)	140			36.4			29.9
Infant birthweight (g)	160	3170	(487)		3089	(442)**	

* $P < 0.05$;** $P < 0.001$.

†Data for all variables not based on same sample of women.

‡Hospital population sample $n = 10\,523$ – $10\,949$.**Table 2.** Proportion agreement, kappa and intraclass correlation coefficients for socio-demographic and obstetric history information evaluated by hospital staff and research project personnel*

Variable	n	Proportion agreement	Kappa‡	Intraclass correlation coefficient	(95% CI)
Maternal education†	140	0.91	0.88		
Husband's education	140	0.87	0.82		
Husband's occupation	140	0.78	0.32		
Total income	140			0.81	(0.74 to 0.86)
House characteristics					
Water (yes/no)	140	0.93	0.72		
Sanitation (yes/no)	140	0.99	0.80		
Floor (type)	140	0.70	0.54		
Television (yes/no)	140	0.74	0.45		
No. persons/room	140			0.75	(0.67 to 0.81)
Obstetric history (abortion, caesarean section, stillbirth)	228	0.96	0.77		
Last pregnancy					
Gestational age	141			0.16	(0.00 to 0.32)
Birthweight	143			0.87	(0.83 to 0.91)
Birth interval (<1 year/>1 year)	146	0.91	0.76		

*Social workers for socio-demographic

†Maternal and husband's education: incomplete primary, complete primary, incomplete high school, complete high school, college.

‡SE of kappa: between 0.04 and 0.08 for all values.

the errors of abstracting and coding perinatal data from medical files have been reported to be very low when trained personnel are used (Hewson & Bennett 1987; Chalmers *et al.* 1976a).

Patients were selected from the clinic at a non-predetermined time by the clinic clerk who was unaware of the nature of the study. Nonetheless,

this process was not completely random and selection bias could have been introduced. However, the sample obtained is very similar to the population of the follow-up study in the distribution of several variables, except for some indication of lower socio-economic status and heavier newborns (80 g) in the study sample

Table 3. Proportion agreement, kappa statistic and intraclass correlation coefficients for antenatal care information collected by hospital staff and the research project resident

Variable	n	Proportion agreement	Kappa*	Intraclass correlation coefficient	(95% CI)
Maternal measurements					
Height	220			0.82	(0.77 to 0.86)
Weight	227			0.99	(0.99 to 1.00)
Uterine height	216			0.92	(0.89 to 0.94)
Blood pressure					
Systolic	221			0.13	(0.00 to 0.25)
Diastolic	221			0.17	(0.04 to 0.30)
Presenting part					
Cephalic/other	154	0.86	0.33		
Oedema (yes/no)	224	0.95	0.12		
Urinary infection	226	0.81	-0.02		
Respiratory infection	226	0.99	-0.02		
Interview					
Gestational age	204			0.98	(0.98 to 0.99)
Pre-pregnancy weight	173			0.95	(0.94 to 0.96)
Smoking (yes/no)	215	0.97	-0.02		
Food supplement	223	0.94	0.74		
Medical treatments (yes/no)	226	0.93	0.82		
Analgesics (yes/no)	226	0.97	0.69		
Tranquilizers (yes/no)	226	0.99	0.50		

* Kappa (SE): between 0.06 and 0.07 for all values.

Table 4. Proportion agreement, kappa statistic and intraclass correlation coefficients for delivery and newborn information collected by hospital staff and the research project resident*

Variable	n	Proportion agreement	Kappa†	Intraclass correlation coefficient	(95% CI)
Oxytocin (yes/no)	127	0.87	0.03		
Anaesthesia (no/yes)	157	0.81	0.71		
Mode of delivery					
(spontaneous/forceps/section)	158	0.98	0.99		
Gestational age (LMP)	156			0.83	(0.77 to 0.87)
Gestational age (physical exam.)	160			0.78	(0.72 to 0.84)
Apgar score					
1 min	160			0.69	(0.60 to 0.76)
5 min	160			0.88	(0.84 to 0.91)
Birthweight (kg)	155			0.96	(0.85 to 0.93)
Length (cm)	154			0.80	(0.73 to 0.85)
Head circumference (cm)	152			0.80	(0.73 to 0.85)

*Variables were collected during labour and immediately after delivery by the research project resident, independently of the hospital records.

†Kappa (SE): between 0.05 and 0.08 for all values.

(Table 1). It seems very unlikely that any selection bias, even if present, would affect the repeated measure values taken by two independent observers.

Several studies have been published recently

on the completeness and accuracy of perinatal data (Greb *et al.* 1987; Doornbos *et al.* 1987; Harter *et al.* 1986; Johnson *et al.* 1985), but only a few reports have evaluated the reliability of data obtained from medical records in clinical

Table 5. Proportion agreement, kappa statistic and intraclass correlation coefficients for physical activity data collected by hospital social worker and research project interviewer

Variable	n	Proportion agreement	Kappa*	Intraclass correlation coefficient	(95% CI)
Overall type of activity (no, physical, other)	140	0.86	0.75		
Help at home (yes/no)	140	0.88	0.72		
Stairs at home (yes/no)	139	0.91	0.61		
Number of blocks walked/day	140			0.43	(0.28 to 0.55)
Work outside home					
Formal paid job (yes/no)	139	0.91	0.82		
Number of hours at work/week	49			0.43	(0.18 to 0.64)
Intensity of activity at work (low/other)	49	0.45	-0.12		
Position at work (seated/standing/other)	52	0.65	0.49		
Stopped working (yes/no)	52	0.12	-0.78		

* Kappa (SE): between 0.06 and 0.13 for all values.

medicine in general (Horwitz & Yu 1984) or in the obstetric literature. (A Medline search was conducted in an attempt to locate and include in this report other perinatal studies of a similar nature.)

Caution should be exercised when interpreting the relation of some variables with a high proportion of agreement but low kappa values. This is due to little data variation and the presence of a rare event.

Approximately 3 months elapsed between the time hospital employees collected data on socio-economic characteristics and physical activity and the time the interviewer collected these data. It is not clear if the lack of agreement for some variables may thus reflect problems with either the patient's recall, the wording of questions or changes in the patient's status over time (Washburn & Montoye 1986).

Some socio-economic characteristics, such as the husband's occupation, the neighbourhood lived in and the number of people in the home, are likely to change among this rather mobile urban poor population. Other variables, such as income, would probably remain similar since it is unlikely that this section of society would have dramatic changes in income in this short time period. Furthermore, if temporal changes in risk characteristics actually do occur during pregnancy, epidemiological studies need to specify and report the time at which data are collected or associations may be obscured and comparisons among studies will be hampered. Similarly, risk

scores using these variables may have to incorporate gestational age at the time of interview to be useful predictors of pregnancy outcome.

Similar to the conclusions from non-pregnant populations (Sallis *et al.* 1985; Washburn & Montoye 1986), data related to work and physical activity during pregnancy tend to have poor reliability. When the questions require detailed description of the work or activity or its intensity, then kappa and intraclass coefficient are low. Unfortunately, it is necessary to obtain this detailed level of information in studies of work during pregnancy, rather than only information on occupational categories (Mamelle & Muñoz 1987).

Obstetric history is perhaps the most important foundation of antenatal care; it helps to identify risk factors and determine care level. The data reported show good reliability for birthweight of the previous infant. This is in agreement with evidence of good maternal recall of significant perinatal events such as birthweight (Seidman *et al.* 1987; Hewson & Bennett 1987; Bennett 1985). This is therefore very important background information, given the strong predictive value of the birthweight history on the present pregnancy birthweight. In contrast, the lack of reliability of gestational age of the previous pregnancy limits its clinical usefulness. Because of the independent influence of previous preterm delivery on the gestational age of the index infant, it is important to improve the

reliability of the reporting of previous gestational age.

Excellent agreement was observed for maternal (Table 3) and newborn (Table 4) anthropometric measures. It is interesting to note that all these clinical measures, except uterine height, were made by trained nurses. The observed results are similar to the high validity reported from a well-nourished population (Ericson *et al.* 1984). The process of standardization including all personnel and the use of same-day reliability test, particularly during the rapid growth of the neonatal period, is likely to have contributed to the high observed reliability values (Donmall 1985).

Poor reliability is exhibited for blood pressure values. This is in agreement with previous published blood pressure data obtained at similar gestational age (MacGillivray *et al.* 1969). In this British study, clinical staff and survey midwives did not show important differences in blood pressure mean values when tested under standard conditions. When the study was repeated in a regular clinic situation, as was the case in our study, relatively large variations were observed between clinic doctors and survey staff (MacGillivray *et al.* 1969). Residency programmes need to emphasize the importance of recording and collecting clinical variables. The measurement of blood pressure should always follow recommended techniques (Petrie *et al.* 1986) and hospitals should provide appropriate equipment within the routine service to avoid unnecessary treatment or follow-up.

Poor agreement was also observed in several other clinical diagnostic factors. In the determination of the presenting part, the poor agreement is due to difficulties in making such a clinical diagnosis as early as 29 weeks gestation. For oedema, urinary and respiratory infection, the lack of uniformly applied criteria could be responsible for the low agreement and accuracy of reporting (Ericson *et al.* 1984).

Gestational age of the index infant, information difficult to obtain in large epidemiological studies, exhibited good reliability when assessed using the date of the last menstrual period or a physical examination of the newborn. The high reliability of this shorter version of the Dubowitz physical examination is consistent with the early report of interobserver agreement of the complete Dubowitz assessment (Dubowitz *et al.* 1970). Effort should be made to include gestational age estimations from physi-

cal examination (short versions) in all perinatal epidemiological studies, after a period of standardization.

We have presented results on the reliability of data collected for a large, prospective perinatal study. Limitation of some variables can be due to methodological problems in phrasing questions, patients' recall, interviewer bias or recording and abstraction of data. We strongly suggest that efforts be made to standardize the questions and instruments carefully. Perinatal studies should always start with a pilot or small reliability study. Furthermore, published results should also include some validity data and examples of the questions used (Gordis 1979). Otherwise it will be difficult to make valid comparisons between studies of work, physical activity, exposure at work place or to generalize their finding.

Perinatal research has become, in the past years, more sophisticated, particularly with the easy availability of computer packages. It should be remembered, however, that the quality of the data remains the backbone of research and that computers will not improve or resolve data quality problems.

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Bone histology and mineral homeostasis in human pregnancy

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Summary. Mineral homeostasis was studied biochemically and histologically in patients in early pregnancy and at term. In early pregnancy there was evidence of increased and reversible resorption of bone, whereas in late pregnancy bone demonstrated active formation and rapid mineralization with minimal resorption. Gut absorption of calcium was not increased in early pregnancy. The overall findings were consistent with calcium liberation from bone in early pregnancy, and with enhanced conservation of bone calcium at term. It is proposed that the additional calcium required during pregnancy is derived largely from the skeleton during early gestation and from dietary absorption at term.

Pregnancy and lactation are conditions which apply stress to calcium and phosphorus homeostasis. In early pregnancy, the rapid increase in maternal plasma volume requires that extra calcium be delivered to the plasma in order to maintain the normality of plasma ionized calcium. In late pregnancy plasma calcium levels must still be maintained despite further increases in plasma volume and the additional drain of mineral being actively transferred from maternal to fetal plasma via the placenta, a process which takes place largely between 28 and 40 weeks gestation (Kumar *et al.* 1980). At term the human fetus contains approximately 32 g of calcium (Cundy & Kanis 1981) which is derived

immediately from maternal plasma and ultimately from the maternal environment. The potential environmental sources of maternal calcium are twofold: mineral may either be withdrawn by bone resorption from the internal or skeletal environment, or it may be withdrawn by gut absorption from the external environment via the diet.

It is known that gut absorption of dietary calcium rises during pregnancy, significant increases being present by 20 weeks (Heaney & Skillman 1971) and it is thought that this process is directly stimulated by the steady increase in circulating 1,25 dihydroxyvitamin D (1,25D) which occurs throughout pregnancy (Whitehead *et al.* 1981). In early pregnancy, the only published study of calcium homeostasis during the first trimester showed that subjects were equally divided between those in positive and in negative calcium balance (Shenolikar 1970).

The contribution made to maternal calcium availability by modification of bone turnover has been little studied. Heaney & Skillman (1971), using the stable isotope ^{45}Ca , were able to calculate that both bone resorption and formation were increased between 30 and 40 weeks gestation and that bone formation alone continued to

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be raised until term. These authors did not study pregnancies before 20 weeks gestation.

Bone behaviour may also be studied using histomorphometric analysis of bone biopsies, and we here present comparative data on the quantitative behaviour of bone specimens taken from healthy patients in early and late pregnancy and from non-pregnant women. The data are complemented by observations on mineral homeostasis and gastrointestinal absorption of calcium in normal pregnant women.

Patients and methods

The 15 patients in the early pregnancy group (mean age 22.4 years) were all between 8 and 10 weeks gestation, and all were scheduled for legal termination of pregnancy. The 13 patients in the late pregnancy group (mean age 25.1 years) were scheduled for elective caesarean section at 39–40 weeks gestation, and all were delivered of healthy singleton infants of appropriate weight for gestation. No pregnant woman had taken calcium or vitamin D preparations during the pregnancy and none suffered from any condition known to affect bone and mineral metabolism. The combined non-pregnant data derive from 14 living and healthy premenopausal women and from autopsy specimens collected previously from 26 normal premenopausal women (overall mean age 33.0 years) who had died suddenly, and who had no history of bone disease or treatment known to affect the skeleton. These patients have been described in detail elsewhere (Aaron *et al.* 1987). All pregnant and non-pregnant women were over 19 years of age, and were deemed to be skeletally mature. The range of ages in each group together with mean and standard deviations are given in Table 1. No living patient had had more than two term pregnancies and the socio-economic background of the groups was comparable.

In each pregnant woman plasma and urine specimens were taken in the fasting state, the

urine specimen being collected 2 h after discard of the first fasting morning specimen.

Plasma total calcium, creatinine, and phosphate were measured on a Technicon SMAC autoanalyser and ionized plasma calcium was determined using a Nova II ion specific electrode. Urinary calcium was measured by atomic absorption spectrophotometry, urinary creatinine and phosphate were all measured by standard autoanalyser techniques and the tubular maximum for reabsorption of calcium (T_mCa) was derived as described by Marshall (1976). Calcium absorption was calculated in fasting patients using oral ^{47}Ca in a 60-min study according to Marshall & Nordin (1981).

Bone biopsy specimens were taken using a trephine with a 0.8 bore diameter, transilial samples of bone being removed from a region 2 cm behind the left anterosuperior iliac spine and 2 cm below the iliac crest. The same operator (D.W.P.) obtained all the biopsies from both pregnancy groups and from the living non-pregnant group.

In preparation for biopsy, and in order to quantify the bone mineralization rate, women in the early pregnancy group and the non-pregnant premenopausal group were labelled with tetracycline by established procedures. Each woman ingested 600 mg of demeclocycline daily for 2 days at 8 and 7 days before the biopsy operation. After removal, biopsies were stained *in vitro* with 1% xylenol orange to provide a second contrasting fluorescent marker (Aaron *et al.* 1984). Specimens were embedded undecalcified in methylmethacrylate (Schenk 1965; Aaron 1976) and 48 serial sections, 8 μ m thick, were cut from each on a Jung K heavy duty microtome. Sections were stained either by the Goldner method (Schenk *et al.* 1969) or in 0.1% toluidine blue, pH 3.5. Established histological features were analysed quantitatively with a Zeiss No. 1 integrating eyepiece and regular histomorphometric point and intersection-counting procedures (Aaron 1976). Measurements included trabecular bone volume as a percentage of the total bony tissue volume in each specimen. Also measured were the surface extent of osteoid borders, the surface extent of resorption cavities relative to total trabecular surface and also in absolute terms whereby intercepts/fields were converted by grid factor to mm^2/mm^3 total tissue (Schenk *et al.* 1969). Calcification fronts relative to osteoid-covered surface were determined and, by means of a calibrated eyepiece

Table 1. Age data in the three groups

Patient group	Age (years)	
	Range	Mean (SD)
Early pregnancy ($n = 15$)	19–29	22.4 (3.0)
Late pregnancy ($n = 13$)	19–34	25.1 (5.0)
Non-pregnant ($n = 40$)	20–48	32.6 (8.1)

graticule, apparent seam width uncorrected for oblique section, and mineralization rate, similarly uncorrected, were calculated. Comparison between the non-pregnant and early pregnancy groups, and between the early and late pregnancy groups was by Student's *t*-test.

In view of the use of a tetracycline and radiocalcium in the early pregnancy group, each patient in this group was seen by two consultants before recruitment so that there should be no doubt as to the firmness of the patient's request for termination of pregnancy. No patient was recruited before a final agreement to perform termination. Subject to the above stipulations ethical approval for this study was obtained from the Leeds West Research Ethics Committee.

Results

The biochemical values obtained in early pregnancy did not differ from normal non-pregnant values save for the fasting urinary calcium/creatinine ratio which was significantly elevated ($P < 0.001$). The differences observed between early and late pregnancy are shown in Table 2. Total plasma calcium was significantly lower at term, but ionized calcium, inorganic phosphate and creatinine showed no change during pregnancy. The fasting urinary calcium/creatinine ratio was significantly lower in late pregnancy, when the fasting urinary phosphate/creatinine ratio was significantly elevated. Tubular maximum for reabsorption of calcium showed a significant rise between early and late pregnancy. Gastrointestinal absorption of calcium, measured only in early pregnancy, was within the normal non-pregnant range.

Histomorphometric analysis failed to show any significant difference between bone biopsies from living non-pregnant premenopausal women and bone removed at autopsy from normal premenopausal women, and these results were therefore combined. Comparison of this group with the group of women in early pregnancy showed a number of histological differences. Similarly there were histological differences in bone between women in early pregnancy and those in late pregnancy (Table 3).

A decline in bone volume in early pregnancy compared with the non-pregnant state was a significant but unforeseen observation. Associated with this was a rise in the relative and absolute surface extent of resorption cavities, and a fall in the relative and absolute extent of surface

osteoid tissue. At the same time, there was no significant difference in seam width, relative extent of calcification fronts nor mineralization rate. In late pregnancy there was a significant increase in bone volume in comparison with early pregnancy. Associated with the recovery of the trabecular bone was a decrease in the extent of resorption cavities to non-pregnant levels and an upward trend in the surface extent of osteoid tissue; the latter, however, while significant in absolute terms, was not statistically significant in relative terms. The relative extent of calcification fronts, high in early pregnancy, was increased further in late pregnancy and there was a marked rise in seam width. The lack of tetracycline labelling in late pregnancy precluded the determination of mineralization rate in this group. However, the reports of a positive relation between seam width and mineralization rate in the absence of osteomalacia (Parfitt *et al.* 1981; Aaron *et al.* 1983) was confirmed here in the biopsies from non-pregnant women and from those in early pregnancy ($r = 0.51$, $P < 0.02$). The increase in seam width in late pregnancy in association with clearly defined and extensive calcification fronts therefore implies an increase in mineralization rate in this group.

Discussion

Our results suggest that the response of the human skeleton to pregnancy is biphasic. Both the bone biopsy results and the increased fasting urinary calcium excretion in early pregnancy suggest that bone resorption is increased at this stage. It was not possible to use urinary excretion of hydroxyproline as a further indicator of bone resorption due to lack of data on its physiological excretion during pregnancy. The significantly lower bone volume in the early pregnancy group compared with both the non-pregnant and the late pregnancy groups, points to rapid, unopposed, and reversible bone resorption at this stage. It is not unusual for histological measurements of resorption rates to appear inadequate with respect to the amount of bone lost over short time periods, as for example in immobilization osteoporosis (Minaire *et al.* 1974) and after oophorectomy (Hodgkinson *et al.* 1978). The increase found in the present study both in relative and absolute terms is not high, and certainly does not resemble that due to massive osteoclastic activity which is encoun-

Table 2. Biochemical variables in early and late pregnancy

Variable	Non-pregnant normal range	Early pregnancy (n = 15)	Late pregnancy (n = 13)
Total plasma calcium (mmol/l)	2.25-2.55	2.36 (0.06)	2.28 (0.04)**
Total ionized calcium (mmol/l)	0.9-1.3	1.18 (0.03)	1.14 (0.04)
Plasma phosphate (mmol/l)	0.8-1.3	1.01 (0.18)	1.04 (0.13) -
Plasma creatinine (μ mol/l)	50-140	60.84 (9.33)	64.63 (9.53)
Urine Ca/creatinine ratio (molar)	<0.15	0.39 (0.04)	0.18 (0.16)**
Urine P/creatinine ratio (molar)	1.0-2.4	0.94 (0.56)	1.60 (0.47)*
T _m Ca (mmol/l GF)	1.8-2.1	1.86 (0.10)	2.19 (0.26)**
Radiocalcium absorption fraction of dose (^{45} Ca)	0.4-1.0	0.71 (0.12)	—

All results are given as mean (SD) values.

Significance of difference between late and early pregnancy groups * $P < 0.005$, ** $P < 0.001$.

Table 3. Histomorphometric variables in the three groups studied

Bone histomorphometry	Non-pregnant controls (n = 40)	Early pregnancy (n = 15)	Late pregnancy (n = 13)
Trabecular bone volume (%)	22.95 (5.93)****	16.15 (4.32)	22.00 (8.08)*
Surface osteoid tissue (%)	5.78 (4.22)**	2.89 (1.94)	5.15 (4.08)
Surface osteoid (mm^2/mm^2)	0.18 (0.15)***	0.07 (0.05)	0.18 (0.19)***
Surface resorption cavities (%)	4.37 (2.02)****	8.07 (3.05)	5.23 (2.80)**
Surface resorption (mm^2/mm^2)	0.14 (0.07)***	0.22 (0.10)	0.16 (0.10)
Osteoid seam width (μ m)	10.11 (3.32)	11.25 (4.84)	16.62 (4.05)***
Calcification front (%)	80.95 (12.33)	86.50 (10.16)	96.42 (5.42)**
Mineralization rate (μ m/day)	0.72 (0.16)	0.56 (0.25)	Not labelled

All results given as mean (SD) values.

Significance of difference between non-pregnant controls and early pregnancy group and between late and early pregnancy groups * $P < 0.05$, ** $P < 0.02$, *** $P < 0.01$, **** $P < 0.001$.

tered in some pathological conditions. However, we are unable to say whether each additional resorption site stimulated by early pregnancy is associated with an increased rate of resorptive activity and this is clearly central to any calculation. No evidence of an osteoblastic response to this enhanced resorption was seen, new bone formation apparently being depressed at this time. Further insight into the precise mechanisms involved may be gained by an examination of the topography of the trabecular bone loss, i.e. whether the remaining trabeculae are simply thinner or are actually fewer in number (Aaron *et al.* 1987).

In late pregnancy, bone formation predominates as is evident both by the histomorphometry and by the significant fall in fasting urinary calcium excretion. The net result of early resorption and later formation would appear to be that there is little net effect on bone mass, since the

bone volume at the completion of pregnancy is similar to that seen in non-pregnant women. This observation is in keeping with previous studies into the determinant of bone mass in older women, which report that pregnancy has either no effect or else a moderate protective influence (Goldsmith & Johnston 1975).

The mechanisms underlying such a biphasic skeletal response to pregnancy are not clear. In particular there is no obvious trigger to the increased resorption which occurs early in pregnancy. This cannot be explained in terms of plasma concentration of 1,25D or parathyroid hormone (PTH) which are the classical modulators of bone resorption. If plasma 1,25D had been elevated this would have been manifest through elevated calcium absorption which was not observed. Indeed this is in keeping with the observations of Bouillon *et al.* (1981) who observed that changes in the circulating con-

centration of 1,25D in early pregnancy are explicable in terms of a pregnancy-induced elevation of D binding protein. Likewise there is no evidence from the literature to support the notion that PTH activity is increased at this stage of pregnancy (Streichen *et al.* 1980).

What evidence there is concerning the concentrations of calcium-regulating hormones in early pregnancy would suggest that resorption should be inhibited due to the presence of elevated circulating calcitonin (CT) (Whitehead *et al.* 1981). This does not exclude the possibility of a humoral stimulation of bone resorption as other agents, such as certain prostaglandins (Conaway *et al.* 1986) and alpha-2HS glycoprotein (Lamkin *et al.* 1987), are now recognized as providing a stimulus to the process, and their activity in early pregnancy is as yet unknown.

Since it is known that the natural history of a bone remodelling unit is for a fairly rapid phase of bone resorption to be followed by a slower period of bone formation during which the bone removed previously is replaced (Frost 1973), it is possible that the increased bone formation occurring late in pregnancy is merely the natural sequel to the early resorption. Indeed, the accepted model of bone turnover requires any bone formation to have been initiated by previous bone resorption (Parfitt *et al.* 1981). It is, however, likely that the bone formation observed late in pregnancy is enhanced by increased circulating levels of total and unbound 1,25D which are present by the third trimester (Bouillon *et al.* 1981; Whitehead *et al.* 1981).

It is therefore possible to speculate on how these changes in bone turnover during pregnancy might arise. Although the fetus itself does not make major demands upon maternal calcium reserves until the last trimester, this phase of gestation together with lactation could be seen as representing a considerable and a prolonged stress upon the mother's skeleton. The changes observed in this study could be viewed as a means of minimizing such adverse effects. Seen in this light the primary purpose of these changes would be that of maximizing bone mass during the final trimester and subsequent lactation. The most important change therefore is the increased bone formation in late pregnancy for which the earlier resorptive phase is an obligatory prerequisite. This might also explain the apparent paradox of calcium being liberated from the skeleton only to be lost in the urine.

Such a hypothesis would predict that the bone

mass achieved at the end of pregnancy would be higher than that at conception. This was not observed in our study where the bone volume at the end of pregnancy was similar to that in the non-pregnant women. On the other hand, this might explain the protective effect of pregnancy on the skeleton observed by some authors (Nilsson 1969). The possible causes of the discrepancy in our study include the fact that the unpaired study design would decrease the sensitivity of detecting such a change and the fact that the non-pregnant and pregnant women were not matched for parity so that the effect of any previous pregnancies upon bone mass might obscure the changes brought about by the present gestation.

Such an interpretation would also offer a possible explanation for osteoporosis of pregnancy. In this condition women experience rapid bone loss and fractures during pregnancy but bone turnover is apparently unremarkable between pregnancies (Smith *et al.* 1985). Such a situation would result if a woman were unable to increase her rate of bone formation to the required level in late pregnancy and would leave her with a considerable skeletal deficit due to the preparative resorption. A similar mechanism might also underlie the heparin-induced osteoporosis which has been described during pregnancy (Avioli 1975). If the drug were to interfere with the response in bone formation, then a picture similar to that of osteoporosis in pregnancy would result.

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Is random plasma glucose an efficient screening test for abnormal glucose tolerance in pregnancy?

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Summary. Random plasma glucose was determined in 276 apparently healthy pregnant women attending our antenatal clinic at 28 to 32 weeks gestation. Mean and standard deviation values within 2 h and >2 h after a meal were calculated. A standard 75-g oral glucose tolerance test was then given to 250 of the 276 pregnant women. Three patients were found to be diabetic and 46 had post-load concentrations indicative of impaired glucose tolerance according to the criteria of the World Health Organization (1980). Using a cut-off point whereby 15% of the population would be tested, we would have identified only 2 of the 3 diabetics and 12 of the 46 with impaired glucose tolerance. This poor predictive power cannot be resolved by altering cut-off points for screening, or by altering the criteria for abnormal glucose tolerance. The basic problem is lack of a close relation between 2-h glucose tolerance value and random glucose when this has been taken >120 min after a meal. In this population with a high prevalence of abnormal glucose tolerance, random plasma glucose is not an efficient screening test.

Screening for gestational diabetes is practised widely on the basis of potential diabetic features in the woman's history, but this may result in 37% (O'Sullivan *et al.* 1973) of the population requiring glucose tolerance tests with a false-negative rate of about one-third (O'Sullivan *et al.* 1973; Gillmer *et al.* 1980). To improve screening the 1-h glucose test has been assessed; this involves measuring plasma glucose 1 h after a 50-g glucose load given without dietary preparation. Taking a cut-off for venous plasma glucose of 7.7 mmol/l resulted in 7.8% of the population being referred for full glucose tolerance test (GTT)

(Gillmer *et al.* 1980), but is likely to have resulted in a 21% false-negative rate (O'Sullivan *et al.* 1973). Lind & Anderson (1984) proposed a simpler screening test which involved measuring a random plasma sample and relating the glucose concentration to the time after the last meal. If the glucose concentration was in excess of the 99% cut-off values for random glucose a GTT was recommended. This appears a very attractive method for a busy antenatal clinic and seems to be gaining in popularity. Before introducing it we decided to assess its sensitivity and specificity in a population with a high prevalence of impaired glucose tolerance, since this has not yet been done.

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Patients and methods

All pregnant women at 28-32 weeks of gestation who were attending one antenatal clinic in the Maternity Hospital of Kuwait and who were not known to be diabetic were asked to participate.

The study was done over an 8-month period. On this basis 282 pregnant women were asked, but six refused to enter the study. In the remaining 276 patients, venous plasma glucose concentrations were measured and the time since the last eating was recorded. All attendances were in the morning, and typical breakfasts are described in the appendix. These 276 patients were given an appointment within 5 days to have a standard 75-g oral GTT. The test was completed by 250 patients. The remaining 26 patients were excluded either because they refused to continue the test (8 patients), data recording was incomplete (6 patients), the plasma glucose report could not be traced (5 patients) or the glucose load was vomited (7 patients). The patients were instructed to be on normal diet for at least 3 days before the GTT and to stop eating and drinking after 22.00 hours on the night before the test. The fasting samples were collected between 08.00 and 11.00 hours the next day. After collection of the fasting samples, each patient drank 75 g glucose in 250 ml of water over the course of 5 min. Venous blood samples were again collected 1 and 2 h after the glucose load into tubes containing sodium fluoride (6 mg/ml of whole blood). Samples were centrifuged and the glucose level in the supernatant plasma was measured by BECKMAN PN 651700 glucose analyser (Beckman Instruments International SA). The criteria for diagnosis are those proposed by the World Health Organization (1980). These criteria define a fasting venous plasma glucose concentration of ≥ 8.0 mmol/l as diagnostic of diabetes. With an oral GTT using 75 g glucose, a venous plasma glucose concentration of ≥ 11.0 mmol/l at 2 h after the glucose load is indicative of diabetes; concentrations between 8.0 mmol/l and 11.0 mmol/l after 2 h indicate impaired glucose tolerance (IGT).

The cut-off points used by Lind & Anderson (1984) were for whole blood which gives glucose values approximately 15% lower than plasma (WHO 1980). Corrected values, of 6.4 mmol/l > 2 h since a meal and 7.0 mmol/l ≤ 2 h after a meal, have been used. Centiles have been calculated from standard deviations.

Results

The mean height of the patients studied was 160.1 cm (SD 5.1) and the mean weight was 73.7 kg (SD 13.1).

Of the 250 patients who had a standard 75-g oral GTT, three were diabetic and 46 exhibited impaired glucose tolerance, an overall rate of 20% for 'abnormal' glucose tolerance. The 276 patients who had a random plasma glucose measurement were divided into two groups according to whether their last meal was within 2 h of sampling. The mean plasma glucose concentrations, standard deviation (SD), and different cut-off values are shown in Table 1. Table 2 shows the predictive power of different cut-off values for the test.

The 276 patients were divided into three main groups according to their nationalities. The first group included all those of Kuwaiti nationality, the second group included all Arabs who were non-Kuwaiti, and the remainder were included in a third group. The mean random plasma glucose concentrations were recalculated for each group (Table 3); there were no statistically significant differences between the nationality groups. The different cut-off values for each group were calculated and used to predict patients with 'abnormal' glucose tolerance. An evaluation of the predictive power of the test using these different cut-off values for each group showed no improvement in performance. Figures 1 and 2 demonstrate that this is not something which can be resolved by changing cut-off points for screening, or by altering the criteria for diagnosing abnormal glucose tolerance. The basic problem is that although there is a reasonable relation between the random plasma glucose value within 2 h of food and the 2-h GTT value ($r = 0.56$), there is a poorer relation between the two variables when the time since last food is > 2 h ($r = 0.22$), and most of the women at our clinic belonged to this latter category.

Discussion

Frank gestational diabetes is known to be common in Kuwait (F. D. Johnstone & A. A. Nasrat, unpublished data). Even with these patients excluded, we found a rate of 20% for 'abnormal' glucose tolerance in an unselected antenatal population. The clinical significance of impaired glucose tolerance in pregnancy using these criteria remains to be clearly defined. Current recommendations that women with impaired glucose tolerance in pregnancy should be managed as if they had diabetes (Watkins 1982) seem clearly inappropriate in Kuwait

Table 1. Random plasma glucose values for 276 patients

Time after last meal (min)	No. of patients	Plasma glucose (mmol/l)			
		Mean (SD)	Cut-off values		
			99%	95%	90%
1-120	95	5.7 (0.96)	7.99	7.30	6.94
≥121	181	4.9 (0.71)	6.54	6.06	5.80

Results are mean (SD) values.

n, Number of patients in each group.

Table 2. The power of two cut-off points for random plasma glucose in predicting a 2-h 75-g oral glucose tolerance of >8 mmol/l

	Lind & Anderson (1984)	90th centile from study population
Percentage of women screened positive	7	15
Sensitivity	0.16	0.29
Specificity	0.96	0.89
Positive predictive value	47%	38%
Negative predictive value	82%	84%
Number of diabetics recognized	1	2
Risk ratio of positive compared with negative random glucose test	×2.7	×2.3

Table 3. Random plasma glucose values by nationality

Time after meal (min)	Plasma glucose (mmol/l)		
	Kuwaiti	Non-Kuwaiti Arab	Others
1-120	5.91 (0.99) (n = 51)	5.64 (1.01) (n = 92)	5.61 (0.77) (n = 36)
≥121	4.96 (0.80) (n = 34)	4.99 (0.74) (n = 46)	4.92 (0.73) (n = 17)

Results are mean (SD) values.

n, Number of patients in each group.

where 1 in 5 of unselected patients has the condition. However, uncertainty will be resolved only by follow-up studies of pregnancy outcome and there is a pressing need for such research in Kuwait. Criteria developed from quite different populations may not necessarily be appropriate for this community. Nevertheless, our basic conclusions from this study are independent of the criteria chosen for defining abnormality.

In an area with high prevalence of gestational diabetes such as Kuwait there are important

obstetric and public health reasons for identifying abnormality, even though the degree of deviation from normal which is clinically significant remains to be clearly defined. Even fairly minor deviations in glucose tolerance in pregnancy seem to be associated with the birth of large babies (Leiken *et al.* 1987) and development of diabetes in the long term (O'Sullivan 1984). Our current criteria for referral for an oral GTT, based on traditional risk factors, mean that more than a third of women should be

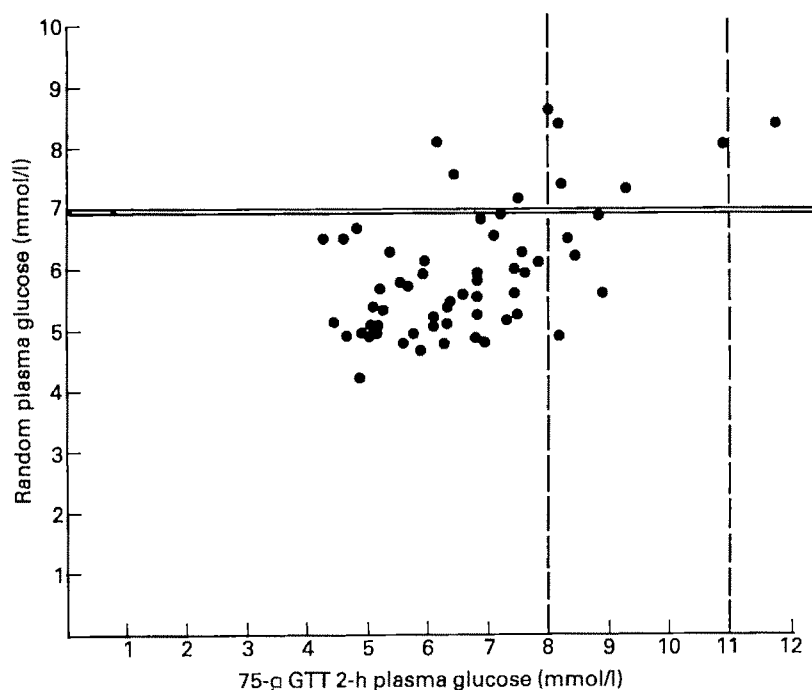


Fig. 1. Relation between random plasma glucose values <120 min after food and the 2-h plasma glucose after the 75-g oral glucose tolerance test (GTT).

The broken vertical lines are the cut-off points recommended by WHO (1980) for plasma glucose 2 h after 75 g glucose indicating IGT and diabetes. The upper horizontal line is the cut-off value for plasma glucose sugar suggested by Lind & Anderson (1984). The lower horizontal line is the 90th centile from our total study population.

referred for testing, but such selection is unreliable. Indeed these risk factors were almost equally distributed in the patients with normal and abnormal glucose tolerance (Nasrat *et al.* 1988). This has been the experience in other populations (O'Sullivan *et al.* 1973; Gillmer *et al.* 1980; Lavin *et al.* 1981). The best documented screening test is the measurement of plasma glucose 1 h after ingestion of 50 g glucose, regardless of time since last meal (O'Sullivan *et al.* 1973). Reed (1984) estimated that 20% of the patients with gestational diabetes will be missed with this original method, and Carpenter & Coustan (1982) modified the method by lowering the test threshold, at the expense of test specificity. Another screening method using HbA_{1c}, either alone or in combination with a 50-g glucose screening test, was insensitive for the detection of gestational diabetes mellitus (Shah *et al.* 1982).

More recently, Lind & Anderson (1984) used random plasma glucose sampling as a screening test. This is simple, cheap, and an extremely attractive practicable method for a busy ante-

natal clinic. Unfortunately, although there is a useful relation between this test and the oral GTT when the woman has eaten within 2 h, this is not so when the duration since eating exceeds 2 h, and this includes most of our patients. Use of the criteria of Lind & Anderson (1984) would allow identification of only 1 of the 3 diabetics and 7 of the 46 women with impaired glucose tolerance. Although a positive screen means that the risk for abnormal glucose tolerance is 1:2.1, a negative screen still carries a risk of 1:5.7. Altering cut-off points or the criteria for abnormal glucose tolerance does not change the basic conclusion that random plasma glucose measurement is of very limited predictive power. A recently published study, comparing plasma glucose at fixed times with 2-h oral GTT values in women selected for glucose tolerance testing, has come to similar conclusions (Jowett *et al.* 1987).

Is random plasma glucose an efficient screening test for abnormal glucose tolerance in pregnancy? Unfortunately not, at least in Kuwait, if

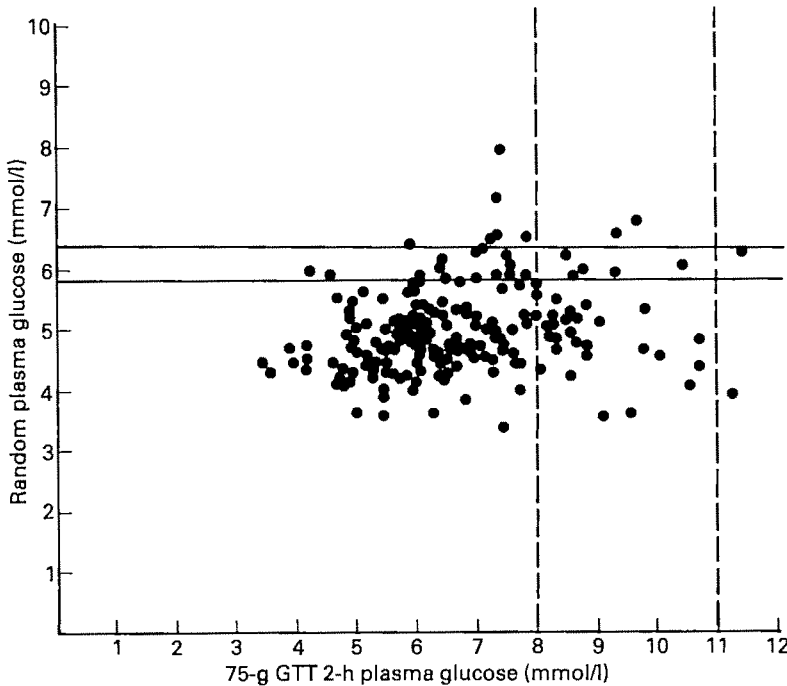


Fig. 2. Relation between random plasma glucose values >120 min after food and the 2-h plasma glucose after the 75-g glucose tolerance test (GTT).

The broken vertical lines are the cut-off points recommended by WHO (1980) for plasma glucose 2 h after 75 g glucose indicating IGT and diabetes. The upper horizontal line is the cut-off value for plasma glucose sugar suggested by Lind & Anderson (1984). The lower horizontal line is the 90th centile from our total study population.

identification of these minor degrees of glucose intolerance is thought to be important.

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Appendix

Dietary survey showed the following to be typical breakfasts in pregnancy: in Kuwaiti women, at least one large piece of bread (arabic or irani), one or two fried eggs, a piece of cheese, and glass of tea with milk and sugar; for Egyptian women, a large piece of arabic or irani bread, a large dish of cooked brown beans with oil and salt, a piece of cheese or one or two boiled eggs, and tea with milk and sugar; for Jordanian and Palestinian women, a large piece of arabic or irani bread, a piece of cheese, olives and tea or coffee with milk and sugar.

Pregnancy complicated by maternal heart disease. A review of 519 women

P. B. McFAUL, J. C. DORNAN, H. LAMKI, D. BOYLE

Summary. Between 1970 and 1983, 519 pregnancies in 405 women with heart disease were managed at the Royal Maternity Hospital, Belfast, Northern Ireland, a rate of 1.3 per 100 deliveries. In 312 (60%) the heart disease was of rheumatic origin, in 161 (31%) congenital, and the remaining 46 (9%) were a miscellaneous group that included arrhythmias, ischaemic heart disease and cardiomyopathies. The New York Heart Association (NYHA) grading was no greater than 1-2 in 445 (86%) pregnancies antenatally. Three maternal deaths occurred, all in the group whose antenatal NYHA grade was 3-4. Heart failure was present in 96 (18%) pregnancies antenatally, and six others developed failure during labour or in the puerperium. Prophylactic antibiotics were not used routinely and infective endocarditis did not occur. The perinatal mortality rate was 19/1000, and the rate of congenital malformations was not raised in the reviewed group.

The steady decline in maternal mortality over the last 20 years has been due largely to a substantial reduction in the number of deaths attributable to hypertensive disorders, haemorrhage, genital sepsis, and abortion (DHSS 1986). The reduction in deaths due to maternal cardiac disease has not been as dramatic and in the most recent enquiry into maternal deaths in England and Wales (DHSS 1986) there were more deaths associated with cardiac disease than with haemorrhage, genital sepsis or abortion.

In this study we present data from 519 pregnancies in 405 women with heart disease who had reached at least 20 weeks gestation. The

study period spans 14 years between 1970 and 1983 during which time the principles governing management were unchanged.

Patients and materials

The notes of all women attending the joint cardiac/antenatal clinic and delivered at the Royal Maternity Hospital, Belfast, between 1970 and 1983 were reviewed. In most women, the cardiac diagnosis had been determined antenatally. Cardiac catheterization had been available throughout the duration of the study and was used to aid diagnosis where there was uncertainty. Echocardiography as a diagnostic tool was available in the last 3 years of the study but was only used in two women to clarify a diagnosis of aortic stenosis. Throughout the study the same cardiologist used the same principles for diagnosis and management.

Visits to the joint clinic run by the cardiologist and an obstetrician were initially every 2 weeks up to 30 weeks gestation and weekly thereafter until delivery. At each visit, a functional grading

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was made according to the criteria of the New York Heart Association (NYHA 1964), and the severity of the cardiac lesion was assessed by detailed clinical examination. Women were advised about the importance of adequate rest, of oral iron supplements and of not smoking. Infection was regarded as serious and was treated with antibiotics. Urine from each woman was screened for asymptomatic bacteriuria and treated if present. Routine admission for bed-rest 1 week before the expected date of delivery was practised where an obstructive or cyanotic heart lesion was present, where there were objective signs that the cardiac status had deteriorated or heart failure had developed. Women with a previous history of endocarditis or those with a prosthetic heart valve were given prophylactic antibiotics during labour. Heart failure was treated with digoxin and diuretics and tachyarrhythmias were treated by drugs (digitalis $\pm$ beta-blockade) alone.

Throughout the period studied, heart disease was not considered to be an indication to induce labour. However, during the period 1976–1979 a unit policy of selective planned delivery was followed in the hospital; this inevitably led to a high induction rate during these years. During the remainder of the study period, labour ward management was influenced by obstetric factors alone. Where labour was induced it was by artificial rupture of the membranes and oxytocin infusion. Forceps were used where normal obstetric indications applied or where there was evidence of heart failure. Pethidine was the analgesic of choice in labour although the use of epidural analgesia was encouraged after it became routinely available in 1980. Intramuscular syntometrine (containing ergometrine maleate 500 mg and oxytocin 5 units) was given immediately after delivery of the anterior shoulder except in patients with heart failure.

Results

Maternal cardiac disease complicated 519 (1.3%) of the 40 970 deliveries at the Royal Maternity Hospital, Belfast, during the 14-year study period.

The main cardiac diagnoses in these pregnancies are given in Table 1. Rheumatic heart disease was twice as common as congenital heart disease overall, but the relative incidence changed and the two categories were about

equally represented in the second half of the period studied.

Most pregnancies (80%) were booked before 20 weeks gestation. Only 5% of the women had a parity greater than five and 80% were aged between 20 and 35 years. Routine elective admission before delivery was not practised, but 20% of the women were admitted antenatally for a cardiac indication. Following delivery 90% were discharged within 10 days. In most pregnancies (85%), the heart disease had been diagnosed before pregnancy, and in the remainder the diagnosis was first suspected at an antenatal clinic before being confirmed by the cardiologist at the joint speciality clinic.

In 312 (60%) pregnancies the cardiac valve lesion was rheumatic in origin. Mitral stenosis was the commonest dominant lesion (61%) followed by mitral regurgitation (33%) and aortic regurgitation (6%).

There were 161 (31%) pregnancies complicated by congenital heart disease (CHD) and the diagnosis and number corrected surgically is given in Table 2. The commonest lesions were septal defects and 70% of the atrial septal defects and 31% of the ventricular septal defects had been corrected. Patent ductus arteriosus accounted for 17% of all CHD and all but one of these had been closed. Pulmonary stenosis and aortic stenosis accounted for 15% and 12% of CHD diagnosis respectively and 33% and 31% had been surgically corrected. There were five women with corrected Fallot's tetralogy. And of pregnancies with cyanotic heart disease there were two women with Eisenmenger's syndrome in which the primary diagnosis was an atrial septal defect. Mitral valve prolapse was not diagnosed during this study and it is only recently that cardiologists have become more aware of this diagnosis. During the study auscultatory findings of an isolated mid-systolic click was considered a normal finding, a late or pan systolic murmur in the mitral area was considered as pathological and due to mitral regurgitation.

Of the 46 pregnancies (9%) grouped together with miscellaneous diagnoses, the most frequent lesions were arrhythmias (Table 3). There were also six pregnancies with ischaemic heart disease, five with secondary pulmonary hypertension, and three with hypertrophic obstructive cardiomyopathy.

Severity of symptoms

Overall, 86% of the pregnancies had a NYHA grade of 1 or 2 antenatally, of these only five

Table 1. Main diagnostic categories of heart disease in 519 pregnancies

Lesion	1970-1983		1970-1976		1977-1983	
	<i>n</i>	(%)	<i>n</i>	(%)	<i>n</i>	(%)
Rheumatic heart disease	312	(60)	195	(76)	117	(45)
Congenital heart disease	161	(31)	52	(20)	109	(42)
Others	46	(9)	12	(4)	34	(13)
Total	519		259		260	

Table 2. Type of congenital heart disease complicating 161 pregnancies

Heart lesion	Pregnancies		Number corrected surgically	
	<i>n</i>	(%)	<i>n</i>	(%)
Atrial septal defect	30	(19)	21	(70)
Ventricular septal defect	29	(18)	9	(31)
Patent ductus arteriosus	28	(17)	27	(97)
Pulmonary stenosis	24	(15)	8	(33)
Aortic stenosis	19	(12)	6	(31)
Coarctation of aorta	18	(11)	14	(78)
Other, acyanotic	2	(1)	0	(—)
Fallot's tetralogy	5	(3)	5	(100)
Congenital heart block	2	(1)	0	(—)
Eisenmenger's syndrome	2	(1)	0	(—)
Primary pulmonary hypertension	1	(1)	0	(—)
Other, cyanotic	1	(1)	0	(—)
Total	161		90	56

deteriorated to grade 3 or 4 in labour or in the puerperium (Table 4). Among the 74 pregnancies whose worst antenatal grade was 3 or 4, 66% improved by the time they were in labour, and in the puerperium only 3% continued to have grade 3 or 4 symptoms. It was within this group of patients with severe symptoms that three maternal deaths occurred.

Cardiac surgery

Of the 405 women in the study 142 had 182 pregnancies after cardiac surgery (Table 5). Mitral valvotomy had been performed in 53 patients, 12 prosthetic heart valves had been inserted and two patients had a graft valve replacement. Of the patients with congenital heart disease 75 previously had some form of corrective surgery. The two largest groups treated surgically were patients with mitral stenosis who had mitral valvotomies, and patients with congenital heart disease corrections. Overall, 45% of mitral stenotic lesions had been treated with valvotomies; of these 73% were grade 1-2 at booking. Of the 55% who had not had surgery 81% were grade 1-2 at booking. In

patients with congenital heart disease 35% had a surgical correction. At booking 98% of these and 89% of uncorrected lesions were graded 1-2 (Table 6).

Mitral valvotomy was performed during pregnancy in two women, both of whom had tight mitral stenosis not previously treated surgically. Both developed severe pulmonary oedema in the second trimester of pregnancy and failed to respond to conventional medical management. Following mitral valvotomies the pregnancies were uncomplicated and healthy infants were born at term.

Delivery

In women with cardiac disease the frequency of assisted delivery was 24%, whereas it was 14% in the overall obstetric population. The caesarean section rate was the same in both groups (13%). There was no difference in the rate of assisted delivery (27%) or caesarean section (13%) between patients with cardiac symptoms grade 1-2 and 3-4.

Table 3. Heart disease: miscellaneous diagnosis

Heart lesion	Number of pregnancies
Supra ventricular tachycardia	10
Tachycardia (others)	7
Ectopic beats	6
Ischaemic heart disease	6
Secondary pulmonary hypertension	5
Hypertrophic obstructive cardiomyopathy	3
Cardiomyopathy (other)	2
Acute myocardial infarction during pregnancy	1
Others	6
Total	46

Cardiac failure

Heart failure was diagnosed if a chest radiograph showed interstitial or alveolar oedema plus one of the following two findings: JVP > 4 cm, or basal crepitations. Failure was present in 96 pregnancies antenatally; 20 (21%) were still in failure during labour and 11 (12%) in the puerperium. In the 423 pregnancies where there was no evidence of heart failure antenatally, one subsequently developed failure during labour and a further five developed failure in the puerperium.

Prophylactic antibiotics

Routine antibiotic prophylaxis against infective endocarditis was given during labour in 3% of pregnancies (previous history of endocarditis or

Table 4. The highest NYHA grade antenatally compared with values during labour and in the puerperium

Antenatal grade	n	(%)*	Grade in labour				Grade in puerperium			
			1-2		3-4		1-2		3-4	
			n	(%)	n	(%)	n	(%)	n	(%)
1-2	445	(86)	440	(99)	5	(1)	440	(99)	5	(1)
3-4	74	(14)	49	(66)	25	(34)	70	(94)	2	(3)
Total*	519		489		30		510		7*	

* Two women died in labour.

Table 5. Type of cardiac surgery performed on 142 women before pregnancy and on two during pregnancy

Operation performed	Before pregnancy n	During pregnancy n
Mitral valvotomy	53	2
Prosthetic valve replacement	12	—
Graft valve replacement	2	—
Correction of congenital heart defect	75	—
Total	142	2

Table 6. Effect of previous cardiac surgery on NYHA grade of symptoms at booking in patients with mitral stenosis or congenital heart disease

Heart lesion	NYHA grade	Previous surgery n (%)	No previous surgery n (%)
Mitral stenosis	1-2	62 (73)	85 (81)
	3-4	23 (27)	20 (19)
	Total no.	85	105
Congenital heart disease	1-2	55 (98)	93 (89)
	3-4	1 (2)	12 (11)
	Total no.	56	105

prosthetic heart valve present). There were no new cases of infective endocarditis.

Perinatal outcome

Overall, 87% of the infants were born at term and the mean birthweight was 3240 g. The perinatal mortality rate (PNMR) in the series was 19/1000 and this compared favourably with a PNMR of 25/1000 among the overall obstetric population during the study period.

Congenital malformations

The frequency of congenital malformations among infants born to mothers with cardiac disease was 5.9% and there was no increase among infants born to mothers with congenital heart disease. This figure is similar to that for our overall obstetric population.

Maternal outcome

The maternal cardiac condition improved or remained unchanged in most patients throughout pregnancy with symptoms deteriorating in just 11.5% of pregnancies (Table 7). There were, however, three deaths accounting for half the maternal mortality in Royal Maternity Hospital during the period:

Patient 1

A 27-year-old primigravida with severe mitral stenosis was transferred to our unit at 28 weeks gestation in congestive heart failure. She had had two mitral valvotomies before this pregnancy. Despite treatment she remained in failure with grade 4 symptoms. At 29 weeks gestation spontaneous onset of labour occurred

but the patient suffered a cardiac arrest and died before the infant was born.

Patient 2

A 34-year-old primigravida with congenital aortic stenosis had ignored medical advice recommending surgical correction before conception. She booked at 18 weeks when grade 4 symptoms and left ventricular failure were noted. Despite admission to hospital and medical treatment her condition did not improve. Surgery was again advised at 20 weeks gestation but was again refused. After the spontaneous onset of labour at 30 weeks gestation, an emergency caesarean section for fetal distress resulted in the birth of a preterm infant with Down's syndrome and severe congenital heart disease who died shortly after birth. The mother was transferred to the intensive care unit where she developed ventricular fibrillation from which she did not recover.

Patient 3

A 24-year-old woman who had one child had pulmonary hypertension secondary to systemic lupus erythematosus and grade 3 or 4 symptoms throughout pregnancy. At no time during pregnancy did she develop heart failure. Labour was induced at 37 weeks because of suspected intra-uterine growth retardation and resulted in the birth of an infant weighing 2130 g. The patient remained well in the puerperium and she was discharged home after 21 days. Three months later she was readmitted to the coronary care unit where she died as a result of severe pulmonary hypertension.

Table 7. A comparison of the cardiac state antenatally and in the puerperium in 519 pregnancies complicated by maternal heart disease

Heart lesion	n	No change or improved		Condition deteriorated		Maternal death	
		n	(%)	n	(%)	n	(%)
Mitral stenosis	190	156	(82)	33	(17)	1	(0.5)
Mitral regurgitation	103	100	(97)	3	(3)	—	—
Aortic regurgitation	19	14	(72)	5	(28)	—	—
Congenital heart disease	161	146	(91)	14	(8)	1	(0.6)
Others	46	40	(87)	5	(12)	1	(2.1)
Total	519	456	(88)	60	(11.5)	3	(0.6)

Discussion

The frequency of pregnancy complicated by maternal heart disease does not appear to have changed over the past 15 years, but recently the relative proportion of rheumatic heart disease appears to have decreased and congenital heart disease increased. This change reflects the increased number of congenital heart lesions that are now treatable surgically and a fall in the frequency of rheumatic carditis. Most pregnancies complicated by maternal heart disease can be expected to have a favourable outcome for both mother and fetus. In this series the perinatal mortality rate (PNMR) of 19 per 1000 births is slightly less than that for the overall obstetric population, illustrating both the remarkably good fetal prognosis in this group and the decline in perinatal mortality rates in general. This has also been reported by others (Sugrue *et al.* 1981; Rush *et al.* 1979), while Batson (1974) and Schaefer *et al.* (1958) found a relatively high PNMR associated with cyanotic heart disease.

Cardiac failure is a serious complication often associated with maternal death (DHSS 1986); it can occur at any time during pregnancy. In this study most cardiac failures occurred in the antenatal period but with appropriate treatment most could be treated allowing labour and the puerperium to be conducted without this added complication. In patients in whom failure was not initially a problem antenatally, it was most likely to develop in the puerperium. We therefore emphasize the need to monitor cardiac patients for heart failure not only antenatally and during labour, but well into the puerperium.

The small group of patients with cyanotic heart disease are important with respect to cardiac and obstetric complications. Eisenmenger's syndrome has been associated with particularly high maternal mortality rates, from 18.7% to 30% in some series (Jones & Howitt 1965; Batson 1974; Loffer 1967; Lindeboom 1948; Gleicher *et al.* 1979). Typical of this syndrome is postpartum syncope and sudden death. Both patients in this study had severe symptoms with evidence of heart failure throughout pregnancy. In each case healthy infants were born with forceps after the spontaneous onset of labour, and no postpartum syncope was experienced.

The commonest type of cardiac surgery performed before pregnancy was a mitral valvotomy. When we looked at the effect of this in

patients originally diagnosed with mitral stenosis it was apparent that surgery does not alone guarantee a complication-free course in subsequent pregnancies. Many of these patients (27%) had severe symptoms at booking. It has previously been noted that the longer the valvotomy to pregnancy interval, the higher the NYHA grading is likely to be (Sugrue *et al.* 1981), suggesting that in general families should be completed with the minimum of delay after valvotomy. We reported only two patients who required valvotomy during the pregnancy. It is our experience that cardiac surgery is only rarely necessary during pregnancy, but when medical treatment has failed, recourse to valvotomy can be undertaken with a good prognosis for the woman and her child.

In many centres it has become established practice to provide antibiotic prophylaxis against infective endocarditis at delivery and in the puerperium for all cardiac patients. Justification for using antibiotics is based on the occurrence of transient asymptomatic bacteraemia in up to 5% of women at delivery (Redleaf & Faddell 1959; Baker *et al.* 1966; Baker & Hubbell 1967) and on reports of endocarditis in the puerperium (Ward & Hickman 1971; Pedowitz & Hellman 1953). The high mortality associated with infective endocarditis certainly calls for every preventive measure in patients at risk. There is, however, no evidence that the long-established antibiotic regimens have an effect on the frequency with which endocarditis occurs following high-risk procedures. In several large series where routine prophylaxis was not given none of the women with rheumatic or congenital heart disease developed endocarditis related to preceding childbirth (Sugrue *et al.* 1980; Smith *et al.* 1976; Fleming 1977). Further support for a reappraisal of antibiotic prophylaxis in cardiac patients around the time of delivery is supported by this report, where no woman developed endocarditis. Moreover, the dual risks of drug toxicity and antibiotic-resistant endocarditis may further complicate the routine use of antibiotic prophylaxis.

In the near future we do not foresee any major changes in the management of pregnancy complicated by maternal heart disease. We would encourage pre-pregnancy counselling by a cardiologist, early diagnosis, shared assessment between cardiologist and obstetrician, attention to detail, continued surveillance well into the

puerperium in order to maximize the chances of a good outcome for mother and baby.

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Comparison of antihypertensive efficacy and perinatal safety of labetalol and methyldopa in the treatment of hypertension in pregnancy: a randomized controlled trial

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Summary. Labetalol was compared with methyldopa in a randomized controlled trial involving 176 pregnant women with mild to moderate hypertension. Diastolic blood pressure below 86 mmHg was obtained in a similar proportion of women given labetalol or methyldopa. Intra-uterine death occurred in four women treated with methyldopa, and the one neonatal death on day 1 occurred in the labetalol group. The average birthweight and the proportion of preterm or small-for-gestational-age babies were similar in both groups. Heart rate, blood pressure, blood glucose, respiratory rate, and Silverman score of the babies did not differ between the two treatment groups, whether the comparison was made for all the infants, or only for those that were preterm or small-for-gestational-age. These data indicate that maternal beta-blockade with labetalol is as safe as methyldopa for the fetus and the newborn.

Methyldopa is the drug most commonly used for the treatment of hypertension in pregnancy and

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is safe for the fetus and the newborn (Redman *et al.* 1976). Although concern has been expressed that beta-blockade may adversely affect fetal growth and neonatal cardiorespiratory function (Dumez *et al.* 1981; Boutroy 1987), maternal ingestion of oxprenolol (Fidler *et al.* 1983; Gallery *et al.* 1985) or labetalol (Lamming *et al.* 1980; Redman 1982) was not associated, in randomized trials, with greater perinatal morbidity or mortality than occurred with methyldopa. However, as these trials involved either low-risk pregnancies with mean birthweights very close to 3000 g (Fidler *et al.* 1983; Gallery *et al.* 1985) or a small number of pregnancies (Lamming *et al.* 1980; Redman 1982), only a few stressed newborns were included in each. Moreover, these reports gave no details concerning the cardiorespiratory effects of methyldopa and beta-

blockers in newborns on variables such as heart rate, blood pressure and respiratory rate.

We compared labetalol and methyldopa for the treatment of mild to moderate hypertension in pregnancy in a randomized controlled trial involving 176 women. Our objective was to compare heart and respiratory rates, blood pressure and blood glucose in babies born in the two treatment groups. Effects on the woman pertinent to perinatal outcome were also carefully assessed, particularly the effectiveness of blood pressure control and the occurrence of proteinuria.

Patients and methods

Twelve maternity clinics participated in this study. Briefly, pregnant women who met eligibility criteria at two baseline examinations were assigned at random to open treatment with either labetalol or methyldopa. Patients were managed both as inpatients and outpatients. The timing and management of delivery were varied to meet the needs of individual patients according to the routine clinical methods of each centre. Mothers and their babies were followed at least to the sixth day after delivery. From the data of Dumez *et al.* (1981) it was possible to estimate that the inclusion of 75 pregnancies in each treatment group would yield a power of 0.95 to detect clinically relevant mean differences of 8 mmHg in blood pressure, 12 beats/min in heart rate and 0.9 mmol/l in blood glucose at a two-tailed alpha of 0.05.

All hypertensive pregnant patients seen in the participating centres between June 1983 and June 1985 were screened for inclusion. Eligibility requirements included a singleton pregnancy, gestational age between 12 and 34 weeks at entry, initial attendance at the participating clinic no later than 20 weeks gestation (menstrual dates) and, on two separate occasions, a diastolic blood pressure exceeding 89 mmHg. Patients with a history of diabetes, rhesus isoimmunization, depression, congestive heart failure, cardiac block or asthma, and patients previously given antihypertensive drugs in the current pregnancy were not included. After giving their informed oral consent, eligible patients were randomly assigned to receive treatment with either labetalol or methyldopa. Randomization, using blinded envelopes, was in stratified blocks of 10 within each clinic. Information obtained at entry included basic demo-

graphic data, medical history, blood pressure, biochemical tests, and fetal ultrasonography. As recommended by the drug manufacturers, starting dosages were 400 mg for labetalol or 500 mg for methyldopa in two daily doses, and dosages were increased as needed to maintain diastolic blood pressure at 85 mmHg or lower. Oral dihydralazine was added if diastolic blood pressure below 86 mmHg could not be obtained with labetalol or methyldopa alone or if large doses of these drugs were poorly tolerated. If the patient was intolerant of the drug used or if the hypertension was refractory, as defined by a diastolic blood pressure higher than 105 mmHg with at least 1200 mg of labetalol plus 100 mg of dihydralazine or 1500 mg methyldopa plus 100 mg of dihydralazine, any treatment could be given and the outcome was assessed on the intention-to-treat basis. Follow-up examination took place every week when diastolic pressure was above 85 mmHg and every 2 weeks when it was below 86 mmHg. Patients were admitted to hospital if the blood pressure exceeded 105 mmHg on treatment, if there was proteinuria, or for other obstetric or fetal indications.

Blood pressure was measured in the sitting position after 5 min of rest. A mercury manometer was used, and diastolic pressure was recorded at the muffling of sounds. Pulse rate was measured by palpation for 30 s after each blood pressure measurement. Proteinuria was considered significant when, on at least two occasions, it was above 1 g/l or a dip-test gave a positive reaction of 2+ or more (Davey & MacGillivray 1986). Ultrasonographic fetal assessment, including measurement of biparietal and abdominal diameters, was performed at entry and then at 32 and 37 weeks. Small-for-gestational-age birthweight was defined as a value less than the 5th centile of expected weight for gestation according to the French distribution (Bréart *et al.* 1977). Neonatal assessment included monitoring of heart rate and blood pressure, measurement of blood glucose, determination of respiratory rate and Silverman score, and was performed at 1, 12, 24, 48 and 72 h after birth. Cardiovascular monitoring used an oscillometric blood pressure monitor with a 2.5 cm wide cuff (Dinamap). Whole blood glucose was measured before feeding on heel prick samples by a glucose oxidase method. In some participating clinics, newborn infants were routinely transferred for 2 or 3 days to a neonatal intensive care unit for monitoring purposes, and determina-

tions at 72 h were omitted for apparently healthy infants. Since the trial was open, pre-natal and neonatal care received in both treatment groups was carefully checked, in order to detect any bias in medical attitudes in connection with a particular treatment.

Data from the randomized groups were compared using Student's *t*-test for quantitative variables and the χ^2 -test for qualitative variables. Yates' correction was applied when the expected number was between 5 and 10, and the Fisher method was applied when the expected number was below 5.

Results

Of 188 eligible patients, 96 were allocated to the labetalol group and 92 to the methyldopa group. Ten patients were lost to follow-up before delivery (three given labetalol and seven given methyldopa) and two patients allocated to treatment with labetalol but started on methyldopa were excluded for protocol violation. Hence, the records of 91 women given labetalol and 85 given methyldopa were analysed. One patient was withdrawn from randomized treatment, but not from follow-up, owing to adverse events; her record was nevertheless included in the analysis of data.

Treatment groups were comparable at entry for demographic characteristics, obstetric history, gestational age, blood pressure, and fetal ultrasonography (Table 1). Prenatal and neonatal care received in both treatment groups is shown in Table 2. No significant difference in maternal attendance to prenatal visits, mode of delivery, or number of newborn infants transferred to an intensive care unit was found between the labetalol and methyldopa groups, but the proportion of infants for whom diastolic blood pressure and blood glucose were recorded tended to be higher in the former group ($P = 0.05$). However, gestational age at birth and birthweight did not differ, in either group, between infants for whom blood pressure and blood glucose were recorded and those for whom they were not available.

Data summarizing maternal outcome are shown in Table 3. The mean highest recorded diastolic pressure as well as mean pressures and heart rates recorded at a given gestational age were very similar in the labetalol and methyldopa groups. However, patients in the methyldopa group were more frequently given

dihydralazine or another antihypertensive drug, or both, than patients in the labetalol group (26% vs 13%, $P < 0.05$, χ^2 -test). In patients given supplementary treatment, mean gestational age, mean systolic and diastolic blood pressures, and the increase in dosage of the initial treatment (median increase: threefold the initial dosage) did not differ between groups at the time when supplementary treatment was added. Side-effects were mild and similar in nature and frequency in both groups. One woman in the labetalol group discontinued treatment because of headache, dizziness and nausea; none in the methyldopa group discontinued treatment. Neither mean plasma urate concentration nor the frequency of significant proteinuria differed between groups. The mode of induction of labour and the frequency of caesarean section were also the same for both groups.

Perinatal outcome is shown in Tables 4 to 6. Four stillbirths occurred in the methyldopa group, of which three were connected with refractory hypertension and severe proteinuria (patients A, B, C in Table 4); one was of unknown origin (patient D). In patient C, labetalol had been added to methyldopa 3 days before delivery because of refractory hypertension. Patient E, allocated to labetalol treatment, gave birth at the 29th week to a baby weighing 1070 g who died 24 h after birth with a bilateral pneumothorax. This pre-eclamptic patient had been given labetalol alone then labetalol plus dihydralazine for 6 days, then, because the diastolic blood pressure remained above 110 mmHg, labetalol was stopped on the seventh day and replaced by intravenous methyldopa and dihydralazine. The pregnancy was terminated by caesarean section 13 days after the patient entered the study and 7 days after labetalol therapy was stopped. For the live-born babies, mean birthweight and gestational age at birth were quite similar in the two groups (Table 5). No significant difference between groups was found for the proportion of preterm or small-for-gestational-age babies, or for the number with an Apgar score of ≤ 7 at 5 min, or those transferred for at least 10 days to a neonatal intensive care unit. Monitoring of heart rate, blood pressure, blood glucose, respiratory rate and Silverman score in the first 48 h of life (Table 6) or at 72 h (not shown) revealed no significant differences between the labetalol and methyldopa groups, whether the comparison was made for all the infants, only for those born

preterm, or only for infants requiring hospitalization for 10 days or more in a neonatal intensive care unit (data from these subgroups will be considered in detail elsewhere).

Discussion

Although antihypertensive treatment has not been shown to decrease perinatal mortality in pregnancies complicated by mild to moderate hypertension (Leather *et al.* 1968; Redman *et al.* 1976; Rubin *et al.* 1983), the administration of various antihypertensive drugs is becoming increasingly a matter of routine. The antihypertensive drug methyldopa is safe for the fetus, newborn, and developing child (Redman *et al.* 1976; Ounsted *et al.* 1980) and is considered the reference drug for treating hypertension in pregnancy. Moreover, early administration of methyldopa in hypertensive pregnancies is associated with a reduction in the frequency of second trimester miscarriage (Leather *et al.* 1968; Redman *et al.* 1976), an effect not necessarily connected with blood pressure control (Redman *et al.* 1976). Since there is only limited evidence for the usefulness of blood pressure reduction *per se* in mild to moderate hypertension in pregnancy, alternative antihypertensive drugs must be carefully compared with methyldopa, especially in terms of perinatal safety. In the case of beta-adrenergic blockers which enter the fetal circulation, special attention should be paid to the potential effects of beta-blockade on the fetus and the newborn, particularly with regard to the risks of impaired fetal growth and of impaired neonatal defence against hypotension, hypoglycaemia and asphyxia (Boutroy 1987; Dumez *et al.* 1981).

Only a few randomized controlled trials have compared a beta-blocker with methyldopa in the treatment of hypertension in pregnancy. In two trials, oxprenolol and methyldopa were compared (Fidler *et al.* 1983; Gallery *et al.* 1985). Neither trial found any difference in mortality between the two treatment groups; one trial (Gallery *et al.* 1985), but not the other (Fidler *et al.* 1983), found that the birthweights were higher in the oxprenolol-treated group. Labetalol was compared with methyldopa in two other randomized trials. Lamming *et al.* (1980) reported that blood pressure was more satisfactorily controlled, that the frequency of proteinuria was lower, and that induced onset of labour was less frequent in 14 patients given

labetalol than in 12 treated with methyldopa. No perinatal death occurred in either group. Comparing 39 patients treated with labetalol with 35 treated with methyldopa, Redman (1982) found no difference regarding maternal blood pressure or proteinuria. At variance with the trial of Lamming *et al.* (1980), he reported induced labour to be somewhat more frequent in the labetalol than in the methyldopa-treated group. Two babies born to women assigned to labetalol died, one on the 3rd and the other on the 34th day, while no perinatal death occurred in the methyldopa-treated group (Redman 1982). In neither trial were any significant differences found in birthweight, gestational age at birth or Apgar score between the two groups.

In accordance with these previous trials (Lamming *et al.* 1980; Redman 1982; Fidler *et al.* 1983; Gallery *et al.* 1985), we found no significant difference in the rate of preterm delivery between women receiving a beta-blocker and those receiving methyldopa. Whereas Gallery *et al.* (1985) reported that there was greater fetal growth in the group treated by oxprenolol than in the group given methyldopa, we found no difference in birthweight or in the number of small-for-gestational-age babies between the two groups. This discrepancy may be explained by differences in methodology, duration of treatment, and study population. Gallery *et al.* (1985) averaged birthweights (as % of normal at a given gestational age) from all pregnancies, whereas in computing the rate of small-for-gestational-age babies, we considered only liveborn babies. They noticed that the difference in fetal growth between oxprenolol and methyldopa diminished with increasing duration of treatment (Gallery *et al.* 1985). Indeed, mean duration of treatment was about 8.5 weeks in the trial described by Gallery *et al.* (1985) as against 12 weeks in our trial. We observed four stillbirths in patients receiving methyldopa, and one neonatal death in a patient initially treated with labetalol but switched to methyldopa before delivery. Although the present trial was not designed to detect differences in perinatal mortality between treatment groups, our data strongly suggest that labetalol is as safe as methyldopa, the reference drug for the treatment of hypertension in pregnancy.

Previously published randomized trials did not report heart rate, blood pressure, or respiratory rate in the newborn babies exposed *in utero* to placentally transferred beta-blockers or

Table 1. Data at entry in two groups of patients

	Labetalol group (n = 91)	Methyldopa group (n = 85)
Maternal characteristics		
Age (years)	27.8 (4.4)	28.9 (6.4)
Height (cm)	162.6 (6.0)	162.6 (5.6)
Weight before pregnancy (kg)	61.3 (10.6)	62.9 (12.2)
Hypertension before pregnancy	29 (32)	28 (33)
Primigravidae	35 (39)	39 (46)
Previous hypertensive pregnancy*	22 (39)	16 (35)
Previous fetal loss*	13 (23)	14 (30)
Previous small-for-date delivery*	6 (11)	8 (17)
Gestational age (weeks)	25.8 (7.5)	26.2 (6.8)
Sitting blood pressure (mmHg)	146/94 (13.4/7.5)	148/96 (13.1/9.0)
Proteinuria >1+	3 (4)	7 (9)
Smokers	8 (9)	6 (7)
Pretreatment ultrasonography		
No. assessed	86 (95)	77 (91)
Growth retardation	9 (11)	7 (9)
Biparietal diameter (mm)	66 (21.0)	61 (23.1)
Abdominal diameter (mm)	68 (17.7)	69 (18.7)

$P > 0.05$ for all comparisons.

*In multigravid patients.

Results show *n* (%) or means (SD) as appropriate.

Table 2. Assessment of maternal and neonatal care in both treatment groups

	Labetalol group	Methyldopa group	<i>P</i>
Maternal care			
No.	91	85	
Hospitalization before term	44 (48)	46 (54)	NS
Onset of labour:			
Spontaneous	40 (44)	35 (41)	NS
Induced	23 (25)	27 (32)	NS
Caesarean section before labour	28 (31)	23 (27)	NS
Caesarean sections (total)	31 (34)	26 (31)	NS
Neonatal care			
No.	91	81	
Admitted to neonatal ICU*	34 (37)	29 (37)	NS
Number with recorded determination of			
Systolic blood pressure	88 (97)	72 (89)	NS
Diastolic blood pressure	88 (97)	71 (88)	=0.05
Heart rate	89 (98)	73 (90)	NS
Respiratory rate	82 (90)	73 (90)	NS
Silverman score	84 (92)	71 (88)	NS
Blood-glucose	88 (97)	71 (88)	=0.05

* Intensive care unit.

Results show *n* (%) of women or infants as appropriate.

methyldopa (Lamming *et al.* 1980; Redman 1982; Fidler *et al.* 1983; Gallery *et al.* 1985). Neonatal blood glucose levels were reported only in one trial (Gallery *et al.* 1979) and were similar in the oxprenolol- and methyldopa-

treated groups. However, it has been reported that neonatal hypoglycaemia may occur as a consequence of maternal beta-blockade (Boutroy 1987). Case-control studies have reported significantly lower blood pressure in babies born to

Table 3. Maternal outcome

	Labetalol group	Methyldopa group	95% confidence interval of difference
Overall maternal outcome			
Total no.	91	85	
Highest diastolic pressure (mmHg)	97.3 (10.9)	98.1 (12.0)	-4.2 to +2.6
Supplementary therapy given	12 (13)	22 (26)	-25% to -1%*
Significant proteinuria†	8 (9)	8 (9)	-9% to +9%
Maximum plasma urate ($\mu\text{mol/l}$)	332 (96)	333 (90)	-28 to +26
Assessment at 30-34 weeks			
No.	84	74	
Sitting blood pressure (mmHg)	133/84 (14/11)	136/87 (14/12)	-7/-7 to +1/+1
Sitting heart rate (beats/min)	83.7 (10.6)	83.3 (9.5)	-2.7 to +3.5
Diastolic pressure >85 mmHg	31 (37)	35 (47)	-25% to +5%
Plasma urate ($\mu\text{mol/l}$)	267 (82)	262 (74)	-19 to +29
Assessment at 35-39 weeks			
No.	78	72	
Sitting blood pressure (mmHg)	131/83 (14/11)	133/85 (16/10)	-7/-5 to +3/+1
Sitting heart rate (beats/min)	82.4 (9.8)	86.2 (12.2)	-7.4 to +0.2
Diastolic pressure >85 mmHg	22 (28)	24 (33)	-20% to +10%
Plasma urate ($\mu\text{mol/l}$)	302 (96)	292 (80)	-18 to +38

* $P < 0.05$, χ^2 -test.†Defined as dip test of $\geq 2+$ on at least two occasions.Conversion: SI to traditional units—Uric acid: $1 \mu\text{mol} = 0.168 \text{ mg}$.Results show means (SD) or n (%) as appropriate.

Table 4. Summary of perinatal deaths

				Treatment (mg/day)		
Gestational age at birth (weeks)	Birth-weight (g)	Proteinuria (g/day)	Highest diastolic (mmHg)	Randomized	At delivery	Duration* (days)
Stillbirths						
A 24	570	4	130	Methyldopa	Methyldopa 1500 Dihydralazine 150	67
B 28	540	4	110	Methyldopa	Methyldopa 3000 Dihydralazine i.v.	36
C 29	850	15	110	Methyldopa	Methyldopa 1500 Labetalol 400†	112
D 28	900	0	100	Methyldopa	Methyldopa 1000	10
Neonatal death						
E 29	1070	3	120	Labetalol	Methyldopa i.v.† Dihydralazine i.v.	13

*In days from inclusion to delivery.

†Labetalol was added to methyldopa 3 days before delivery in patient C, and methyldopa was substituted for labetalol 7 days before delivery in patient E, due to refractory hypertension (see text).

women treated with a beta-blocker than in those born to women treated with methyldopa (Dumez *et al.* 1981; MacPherson *et al.* 1986). Furthermore, the former had a lower heart rate than the latter (Dumez *et al.* 1981). Discrepancies between the present trial and such studies

could be due to differences in power. In the present study, 91 women treated with labetalol and 85 treated with methyldopa were compared, as were 91 and 81 liveborn babies exposed to the transplacental passage of labetalol and methyldopa, respectively. Clinically relevant differ-

Table 5. Data for liveborn babies in both treatment groups

	Labetalol group	Methyldopa group	95% confidence interval of difference
Total no.	91	81	
Gestational age (weeks)	37.8 (2.6)	37.8 (2.5)	-0.8 to +0.8
Birthweight (g)	2860 (728)	2897 (727)	-255 to +181
Small-for-gestational-age	11 (12)	12 (15)	-13% to +7%
Born before 37 weeks	22 (24)	17 (21)	-9% to +15%
Apgar score <8 at 5 min	10 (11)	5 (6)	-3% to +13%
Intensive care unit >9 days	20 (22)	18 (22)	-12% to +12%

$P > 0.05$ for all comparisons.

Results show means (SD) or n (%) values as appropriate.

Table 6. Mean heart rate, blood pressure, blood glucose and respiratory rate, and proportion with a Silverman score above 0 in liveborn babies

	Labetalol group	Methyldopa group	95% confidence interval of difference
Heart rate (beats/min)			
1st hour	(67) 135	(57) 137	-8.2 to +4.2
12th hour	(81) 129	(67) 131	-7.5 to +3.5
24th hour	(83) 134	(67) 132	-2.9 to +6.9
48th hour	(79) 133	(65) 132	-4.1 to +6.1
Blood pressure (mmHg)			
1st hour	(59) 69/39	(44) 68/41	-5.0/-7.4 to +7.0/+3.4
12th hour	(79) 71/42	(63) 72/43	-6.5/-5.7 to +4.5/+3.7
24th hour	(83) 73/45	(69) 77/47	-9.0/-6.4 to +1.0/+2.4
48th hour	(79) 76/48	(65) 80/52	-9.9/-9.6 to +1.9/+1.6
Blood glucose (mmol/l)			
1st hour	(34) 3.12	(26) 3.64	-1.4 to +0.4
12th hour	(65) 2.88	(60) 3.26	-0.8 to +0.1
24th hour	(82) 3.12	(61) 3.38	-0.7 to +0.2
48th hour	(71) 3.21	(62) 3.42	-0.6 to +0.2
Respiratory rate (/min)			
1st hour	(61) 48.3	(55) 48.9	-4.8 to +3.6
12th hour	(69) 49.6	(60) 48.4	-2.9 to +5.3
24th hour	(68) 45.9	(59) 47.6	-4.5 to +2.1
48th hour	(66) 44.3	(60) 46.7	-6.5 to +1.7
Silverman score >0			
1st hour	(66) 18%	(56) 16%	-11% to +15%
12th hour	(72) 4%	(59) 9%	-14% to +4%
24th hour	(75) 3%	(62) 3%	-6% to +6%
48th hour	(73) 0	(63) 0	

$P > 0.05$ for all comparisons.

The number of newborn infants assessed at a given time is shown in parenthesis.

ences of 7.5 mmHg in diastolic pressure, 8.5 beats/min in heart rate and 0.8 mmol/l in blood glucose between newborns of each group could be detected at the alpha and beta risks of 0.05, i.e. differences similar to MacPherson *et al.* (1986) or half as great as Dumez *et al.* (1981). Given the power of our trial and that the differ-

ences we observed between the two treated groups were close to zero, the hypothesis that labetalol has serious adverse cardiorespiratory effects may be rejected, at least for patients similar to those studied here. Differences in results could also be due to the characteristics of the study population: newborn infants studied in

the present trial were perhaps at lower risk than those reported in other trials (Dumez *et al.* 1981; MacPherson *et al.* 1986; Boutroy 1987). However, in our trial there were more than 20 pre-term or small-for-gestational-age infants in each group, and no significant difference was found to be associated with treatment in these high-risk subgroups. Finally, differences between this trial and reports about the use of acebutolol (Dumez *et al.* 1981) or other beta-blockers (Boutroy 1987) could be explained on pharmacological grounds. Labetalol is a beta-blocker with alpha adrenoceptor antagonist properties which limit the bradycardia induced by beta-blockade and may cause peripheral vasodilation (Louis *et al.* 1984). Indeed, we saw no difference in maternal heart rate between the two groups. Moreover, labetalol has been shown to decrease maternal blood pressure and placental vascular resistance and to pass the placental barrier with no alteration in Doppler ultrasound indices of fetal aortic blood flow (Jouppila *et al.* 1986). Furthermore, it must be emphasized that the neonatal effects observed with a given beta-blocking agent cannot be extrapolated to other beta-blockers since these drugs differ not only in their pharmacodynamic effects but also in their transplacental availability to the fetus.

The similar perinatal outcome observed in the labetalol and methyldopa groups was mirrored by the effects on the mothers. At variance with Lamming *et al.* (1980), we found no difference in the frequency of proteinuria in women treated with labetalol or methyldopa. As regards maternal blood pressure, the adoption of a large dose range for both drugs and the availability of supplementary treatment—mainly dihydralazine—allowed the target diastolic pressure of 85 mmHg to be reached in most of the pregnancies. Given the design of the protocol, neither mean diastolic blood pressure in weeks 30–34 and 35–39 of pregnancy nor the highest diastolic pressure recorded differed significantly in the labetalol and methyldopa groups. Note, however, that blood pressure control was more frequently achieved with labetalol alone than with methyldopa alone. It may be argued that in an open study, this difference could be due to the bias arising from the physician's preference for the dosage of labetalol and methyldopa that he or she considers to be effective and well tolerated. However, the fact that supplementary treatment was added at similar gestational ages and blood pressure levels, i.e. to equipotent

doses of methyldopa and labetalol, does not support this argument. Labetalol may be a more convenient first-line antihypertensive agent than methyldopa.

In conclusion, the present trial was designed to compare the antihypertensive effects and perinatal safety of labetalol and methyldopa in the treatment of mild to moderate hypertension in pregnancy. At doses ensuring blood pressure control in similar proportions of pregnancies, the two treatments were found to be equally safe for the newborn. It must be emphasized, however, that the question of which hypertensive pregnant women should be treated and at what blood pressure level remains unsettled. Since no published trial has shown any maternal or perinatal benefit from antihypertensive drugs other than a decrease in blood pressure (Leather *et al.* 1968; Redman *et al.* 1976; Rubin *et al.* 1983) leading to less frequent maternal hospitalization (Rubin *et al.* 1983), the reversal by antihypertensive treatment of the perinatal consequences of hypertension in pregnancy remains hypothetical. Any decision to give antihypertensive drugs should have maternal objectives: to ensure maternal safety when the diastolic blood pressure is above 110 mmHg (Redman *et al.* 1976), or to ensure the continuity of preventive care in women treated before pregnancy for permanent hypertension. For these purposes, labetalol can be considered a safe and convenient alternative to methyldopa.

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Human leucocyte antigen status in African women with eclampsia

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Summary. Investigation of the HLA system in 53 African eclamptic or imminently eclamptic women showed that they were significantly more likely to be heterozygous at the B locus than were normal controls. This did not apply to the A or D related loci.

The aetiology of eclampsia remains unknown, but suggestions that it may be an immunogenetic disorder date back to the beginning of the century (Beer & Need 1985). Redman *et al.* (1978) reported that women who had only one detectable surface antigen determined by the HLA-B locus were at an increased risk of developing pre-eclampsia in pregnancy. This work has yet to be confirmed. To examine this association, we studied the HLA system in women who developed eclampsia and compared it with that found in the normal population.

Patients and methods

The study included 53 black African women from Zululand admitted to the labour ward of King Edward VIII hospital who had systolic blood pressures greater than 160 mmHg and diastolic blood pressures greater than 115 mmHg, gross oedema and at least 2+ proteinuria by standard turbidimetric methods in a catheter specimen of urine. Forty-two patients had already suffered at least one seizure before

admission and the remaining 11 all complained of headache, nausea and visual disturbances. All were either irritable or had intellectual clouding and an independent observer described those who had not had a seizure as being hyper-reflexic with clonus before therapy. Patients with a history of neuropathology, diabetes, hypertension, renal disease, recurrent miscarriage or a recent blood transfusion were all excluded. Patients who were still proteinuric or hypertensive ($>140/90$ mmHg) 12 days after delivery were also excluded. Three patients with eclampsia did not know their parents, but the remainder denied that they could be the product of consanguineous marriage or matings.

The control group for the A and B locus of the HLA system consisted of 1416 blood donors of the same tribe and resident within the same hospital catchment area and 412 of them also acted as controls for the DR locus.

The HLA, A, B and DR antigens were determined by a two-stage lymphocytotoxicity test (Terasaki & McClelland 1964). Patients with only one antigen per locus were considered to be homozygous at that locus. Frequency differences between the eclamptic patients and the controls were tested for significance with the χ^2 -test. The formula given by Haldane (1956) was used to combine data from the available published series with the data presented. Relative risk was defined as the number of times more often the disease occurred in women positive for that antigen than in those negative for that antigen (Woolf 1955).

Results

Clinical details of the subject group are recorded in Table 1. Women with eclampsia or imminent

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Table 1. Clinical details of patients with eclampsia

Variable	Number
No. of patients	53
No. with previous viable pregnancy	26 (49%)
Nulliparous	27 (51%)
Preterm labour (< 35 weeks gestation)	26 (49%)
Age (years)	
< 20	16
20-24	21
25-30	11
> 30	5
Maternal deaths	1*

*Intracranial haemorrhage.

eclampsia were less likely to have only one detectable antigen at the B locus than were the normal population, the difference was statistically significant ($P < 0.01$, $\chi^2 = 7.4$). The relative risk in patients in whom both antigens were detected at the B locus is thus increased to 2.3. This does not apply to the A locus or at the DR locus (Table 2). No specific A, B or DR antigen occurred more commonly in eclamptic patients.

Discussion

Two antigen types are inherited, one from each parent. If only one antigen can be detected, then it can be inferred that the individual has either inherited the same antigen from each parent, thus making her homozygous at that locus, or that she has an antigen yet to be discovered. As it is believed that over 98% of the B locus antigens are known to us, the finding of a single B locus antigen is presumed to be synonymous with homozygosity. Pregnant Zulu women suffering from eclampsia or imminent eclampsia are less likely to have only one detectable human lymphocyte antigen at the B locus than are the normal, healthy population from the same tribe and district. Our eclamptics are more likely to be heterozygous at the B locus. It is perhaps signifi-

cant that women born of a consanguineous relationship, and thus relatively homozygous, have some protection from developing eclampsia in pregnancy (Stevenson *et al.* 1976). However, heterozygosity in eclamptics does not occur at the A or DR locus, this observation is unlikely to be related.

Our data contradict the findings of Redman *et al.* (1978). They studied 80 Oxfordshire women suffering from pre-eclampsia and computed a P value of 0.025 supporting an association between pre-eclampsia and homozygosity at the B locus. Simon *et al.* (1980) also reported that French pre-eclamptic patients were relatively homozygous, but they only recruited 26 patients (six were homozygous) and their control group was limited to 16 men, none of whom was homozygous. However, Persitz *et al.* (1983) investigated 40 women in Israel, and Scott *et al.* (1976) studied 46 women from Iowa with eclampsia and pre-eclampsia and both studies failed to show such a relation. It is difficult to understand why English pre-eclamptic patients should tend towards homozygosity at the A and more particularly at the B locus yet Africans with eclampsia are more likely to be heterozygous. An immunogenetic explanation seems unlikely. It is true, however, that some disorders are associated with an HLA type only in certain races, e.g. HLA-B54 is associated with juvenile diabetes mellitus in Japanese, but not in Caucasians. No condition yet described is associated with such marked polarization as demonstrated here. If the explanation of such conflicting results is not within the different racial study groups then it may be with the disease. We have presumed that pre-eclamptics become eclamptics and therefore the two groups are comparable. However, patients presenting with acute eclampsia may have a different genotype compared with those presenting with proteinuric hypertension in pregnancy. Finally, it must be noted that 30% of our control population are

Table 2. Frequency of human leucocyte antigen (HLA) homozygosity in patients with eclampsia and in normal controls

HLA locus	Eclamptic patients		Normal controls	
	<i>n</i>	(%)	<i>n</i>	(%)
A	19/53	(35.8)	374/1416	(26.4)
B	7/53	(13.2)	435/1416	(30.7)*
DR	22/53	(41.5)	223/412	(54.1)

Significance of difference between the two groups. * $P < 0.01$, $\chi^2 = 7.4$.

homozygous. This remarkably high figure may be a reflection of African tribal society and the immobility of its members due to political and economic constraints. Such nuclear communities are not a feature of Oxfordshire, Iowa or France.

The history of HLA associations with certain diseases has been a major breakthrough in our understanding of the genetics of many diseases. The exciting work by Redman *et al.* (1978) associating pre-eclampsia with homozygosity at the B locus promoted many ideas and strengthened the immunogenetic interest in the subject. It is not clear why in African women we found a significant association between heterozygosity and eclampsia, just the opposite of what we expected. If all the available literature is gathered it conflicts and when it is summated no trend emerges. Eclampsia is either independent of HLA status or the association is so complicated it defies present comprehension.

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Thyroid function in pre-eclampsia

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Summary. Thyroid function was studied in 24 proteinuric pre-eclamptic patients and 24 gestation-matched controls. The pre-eclamptic patients had significantly lower mean total (TT4) and free thyroxine (FT4), and thyroxine-binding globulin (TBG) concentrations, and the concentration of thyroid-stimulating hormone was significantly higher. The mean total triiodothyronine (TT3) concentration, however, was similar to the controls. This is at variance with the classical low T3 syndrome reported previously for pre-eclamptic patients. Compared to the controls, 33.3% and 29.2% of the pre-eclamptic patients had low TT4 and TT3 concentrations respectively. Pre-eclamptic patients with low TT4/TT3 had significantly higher plasma urate concentrations. Thyroid hormone concentrations may reflect the severity of pre-eclampsia.

In normal pregnancy, changes in thyroid function are well documented (Smith & Bold 1983; Franklyn *et al.* 1983; Parker 1985), but information about thyroid function in complicated pregnancies is scanty. Hypertension and related complications have been reported in association with thyroid disorders, including postpartum thyroiditis (White & Andreoli 1984, 1986) and thyrotoxicosis (Menon *et al.* 1982). This association may not be coincidental, since thyrotoxicosis may induce hypertension (Fowler 1982). On the other hand, high concentrations of total and free thyroxine (TT4 and FT4) and subnormal triiodothyronine (TT3) and free triiodothyronine (FT3) concentrations have been reported in pre-eclamptic patients (Osathanondh *et al.* 1976; Tolino *et al.* 1985). In a patient who had eclampsia, TT4 and FT4 concentrations were low and TT3 and FT3 were undetectable (Osathanondh *et al.* 1976). In view

of these reports we have re-examined thyroid function in pre-eclamptic and normal pregnant women in the third trimester of pregnancy.

Patients and methods

The study included 24 women with proteinuric pre-eclampsia and a parity-matched control group recruited from normotensive antenatal patients. Pre-eclampsia was diagnosed in hospital at a blood pressure of $\geq 140/90$ mmHg on two or more occasions at least 6 h apart. Proteinuria was diagnosed when a reading of $\geq 1+$ on the dipstick was found in repeated clean-catch midstream urine samples. None of the pre-eclamptic patients or the controls had clinical features of thyrotoxicosis or was on medication that might affect her thyroid function. For this study, blood was taken from the pre-eclamptic patients in hospital after the diagnosis was made and before delivery. Blood was taken from each control subject either at the antenatal clinic or in hospital before labour or elective caesarean section at the same gestation as the matched study subject.

Plasma was separated as soon as possible and stored at -20°C until assayed for thyroid hormones. Plasma TT4 was measured by an enzyme multiplied method (SYVA EMIT) adapted for

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use on the Cobas-Bio centrifugal analyser. Plasma FT4, TT3, FT3 and thyroid-stimulating hormone (TSH) were measured by radioimmunoassay and T3 uptake (T3U) by radioassay using commercial kits from Diagnostic Product Corporation. Plasma thyroxine-binding globulin (TBG) was measured by the Corning radioimmunoassay kit. Statistical comparison was by Student's *t*-test and χ^2 -test where appropriate.

Results

There were four parous patients in each group. None of the pre-eclamptic patients had a history of renal disease or abnormal renal function tests before or after delivery. The mean gestation of the two groups at presentation and blood sampling was 37.3 weeks (range 31–41 weeks). There was no difference in the mean age between the two groups (Table 1). The pre-eclamptic patients were delivered at an earlier gestation ($P<0.01$), and had lower infant birthweights ($P<0.01$) than the control group

although there was no difference in the mode of delivery. Five of the ten caesarean sections in the pre-eclamptic patients were performed for suspected fetal distress, including one with placental abruption. In the control group only two caesarean sections were for suspected fetal distress. There were no serious neonatal complications in either group, and all babies were subsequently discharged alive and well.

Thyroid function in the two groups is shown in Table 2. In both groups the mean values of thyroid hormones, except for T3U, were within the normal laboratory reference ranges. Pre-eclamptic patients had significantly lower mean TT4, FT4 and TBG concentrations, and significantly higher TSH concentrations compared to the controls. The free thyroxine index (FTI) was slightly but not significantly lower in the pre-eclamptics.

Nine pre-eclamptic patients had small-for-gestational-age (SGA) babies, birthweight $\leq$ the 10th centile. They had lower mean concentrations of TT4, FT4, FT3 and FTI, and higher mean TT3 and TBG concentrations than

Table 1. Obstetric outcome in pre-eclamptic patients and controls

	Pre-eclamptic patients (<i>n</i> = 24)	Controls (<i>n</i> = 24)	Difference
Age (years)	28.4 (5.2)	27.5 (5.1)	NS
Delivery			
Gestation (weeks)	37.5 (2.3)	39.3 (1.4)	$P<0.01$
Mode			
Vaginal	14	17	NS
Caesarean section	10	7	
Birthweight (g)	2750 (650)	3240 (420)	$P<0.01$

Results are mean (SD) values.

Table 2. Thyroid function in pre-eclamptic patients and controls

	Normal laboratory range	Pre-eclamptic patients (<i>n</i> = 24)			Controls (<i>n</i> = 24)			<i>t</i> -test
		Mean	(SD)	(range)	Mean	(SD)	(range)	
TT4 (nmol/l)	46–140	114.0	(27.4)	(60–175)	131.6	(23.2)	(106–187)	$P<0.05$
TT3 (nmol/l)	1.0–3.1	2.0	(0.9)	(0.9–4.5)	2.1	(0.5)	(1.4–2.9)	NS
FT4 (pmol/l)	7–21.8	12.4	(4.6)	(7.2–26.7)	16.3	(4.4)	(9.2–27.8)	$P<0.01$
FT3 (pmol/l)	2.2–6.3	2.9	(1.1)	(0.9–5.9)	3.3	(0.8)	(2.0–5.4)	NS
TBG (mg/l)	11.5–32.2	37.4	(8.8)	(27.6–64.7)	43.5	(7.8)	(27.1–65.5)	$P<0.02$
T3U (%)	24–38	22.1	(2.6)	(19–28)	20.7	(2.3)	(16–26)	NS
FTI		24.8	(6.6)	(13.8–47.3)	27.0	(4.4)	(20.3–36.1)	NS
TSH (m.i.u./l)	0.4–5.2	3.7	(1.3)	(1–7.3)	2.5	(0.8)	(1–5.2)	$P<0.001$

the other pre-eclamptic patients with appropriate-for-gestational-age (AGA) babies (Table 3). However, none of these differences was statistically significant.

In the pre-eclamptic patients, eight had TT4 concentrations below the range in the controls, including five with low TT3. Another two pre-eclamptic patients had low TT3 only. When these 10 pre-eclamptic patients were compared to the others with normal TT4 and TT3 concentrations, there was no difference in the highest mean arterial pressure before treatment or delivery, or in the platelet count (Table 4). However, these 10 patients had a significantly higher mean concentration of plasma urate ($P < 0.05$), and the mean gestational ages at delivery and birthweight were lower, although the differences were not statistically significant. This group also had twice the rate of SGA babies, but this difference was not statistically significant.

Discussion

Our results suggest that pre-eclamptic patients have lower concentrations of both TT4 and FT4, and higher concentration of TSH, at the time of

diagnosis. In our pre-eclamptic patients, 33.3% (8/24) had low TT4, and although TT3 concentration was similar to that in the control group, 29.2% (7/24) had TT3 values below the range found in normal pregnant controls. Our findings of low TT4 and FT4 at the time of diagnosis of pre-eclampsia were, however, at variance with the two previous reports, each with 10 patients, in which the TT4 and FT4 concentrations were higher than the controls (Osathanondh *et al.* 1976; Tolino *et al.* 1985). Despite a significantly lower TBG level in our pre-eclamptic patients, and the reported correlation between FT4 level (using analogue method) and albumin concentration (Sakata *et al.* 1986) which might have been low in pre-eclamptic patients, we think that the observed low TT4 and FT4 concentrations are a genuine feature because the TSH concentration in pre-eclamptic patients was significantly higher, and the free thyroxine index was also slightly but not significantly lower in the pre-eclamptics. Thus the hypothyroxinaemia may suggest an element of hypothyroidism in these patients, since pregnancy is generally associated with hyperthyroxinaemia (Smith & Bold 1983). As the low T3

Table 3. Thyroid function in pre-eclamptic patients—relationship to birthweight

	Small-for-dates (<i>n</i> = 9)	Appropriate-for-dates (<i>n</i> = 15)
TT4 (nmol/l)	106.0 (30.8)	119.1 (23.2)
TT3 (nmol/l)	2.10 (1.2)	1.9 (0.6)
FT4 (pmol/l)	11.6 (4.1)	12.9 (4.7)
FT3 (pmol/l)	2.8 (1.2)	3.0 (1.0)
TBG (mg/l)	39.5 (12.1)	36.2 (5.6)
T3U (%)	22.0 (2.7)	22.3 (2.6)
FTI	22.8 (5.4)	26.1 (7.3)
TSH (m-i.u./l)	4.3 (1.4)	3.3 (1.0)

Results are means (SD).

Table 4. Severity of disease in 24 pre-eclamptic women in relation to concentrations of TT4 and TT3

	Low TT4 or TT3 levels (<i>n</i> = 10)	Normal TT4 or TT3 levels (<i>n</i> = 14)
Mean arterial pressure (mmHg)	121.0 (11.7)	121.5 (9.6)
Plasma urate (mmol/l)	0.50 (0.13)	0.36 (0.13)*
Platelet count ($\times 10^9/l$)	180.5 (61.7)	173.4 (54.2)
Gestation at delivery (weeks)	36.8 (2.6)	38.0 (2.1)
Birthweight (g)	2470 (610)	2888 (549)
Number with SGA babies (%)	5 (50%)	4 (28.6%)

**t*-test $P < 0.05$.

Results are mean (SD) values.

syndrome occurs in situations such as protein-caloric malnutrition and other systemic diseases (Cavalieri & Rapoport 1977), the classical features would probably only be found in women with severe pre-eclampsia at a later stage. The differences in our observation may therefore be accounted for by the different time of blood sampling in relation to the duration and stage of pre-eclampsia. Our recent observation of a marked drop in TT3 concentration within 4 days after delivery in a pre-eclamptic patient carrying a hydropic fetus (Lao *et al.* 1988) tends to support this hypothesis. However, we cannot compare our results with the two previous studies because in these reports the time of blood sampling in relation to gestation at presentation and duration of pre-eclampsia was not elaborated on.

Our results also suggest that the degree of hypothyroxinaemia might reflect the severity of pre-eclampsia. Our pre-eclamptic patients with SGA babies appeared to have a greater degree of hypothyroxinaemia and higher TSH concentration. The lack of a statistically significant difference may be due to the small number of patients in the study. In addition, the patients with low TT4 or TT3 had significantly higher plasma urate concentration and double the rate of SGA babies. The changes in thyroid hormones in complicated pregnancies therefore merit further studies to see if they reflect the severity of the conditions.

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Poor maternal weight gain between 28 and 32 weeks gestation may predict small-for-gestational-age infants

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Summary. In a retrospective analysis of 158 women considered to have had normal, low-risk pregnancies, 30 gave birth to infants with a birth-weight less than the 10th centile for gestation. These 30 women had a significantly poorer mean increase in weight (0.99 kg) between 28 and 32 weeks gestation than the other 128 women (1.95 kg) who gave birth to infants with birthweights above the 10th centile for gestation. There was no statistically significant difference in booking weight, overall weight gain or other variables associated with low birthweight between the two groups of women which suggests that poor maternal weight gain specifically between 28 and 32 weeks gestation may predict small-for-gestational-age infants

An important objective of antenatal care is the identification of babies who may be small-for-gestational-age (SGA) early enough to influence the outcome of the pregnancy. Screening procedures, whether clinical, biochemical or using ultrasound, fail to detect a significant proportion of these infants and have substantial limitations because of expense, clinic time or inconvenience to patients.

Most antenatal practices rely on the woman's obstetric or medical history, socio-economic status or complications in the present pregnancy to identify infants with an above average risk of

being born SGA. This approach may permit intensive, hospital-based supervision of these patients, but only 50% of growth-retarded babies are identified antenatally (Low & Galbraith 1974; Rosenberg *et al.* 1982). In addition, false-positive pregnancies may be inappropriately assigned to a high-risk clinic.

A clinical observation that three consecutive stillbirths in this hospital were born to mothers in a 'low-risk' category who had zero weight gain between 28 and 32 weeks, but a normal overall weight gain, prompted us to investigate the possibility that poor maternal weight gain during the early third trimester may indicate an increased risk of giving birth to a SGA infant and therefore at risk from any associated complications.

Patients and methods

The case records of 334 consecutive deliveries at Wythenshawe Hospital over a 6-week period were studied retrospectively. The population of the area is predominantly white European and most women booking for confinement at the hospital are offered a routine ultrasound examination around 16-18 weeks which can be used to

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check the gestation. Only women who were delivered between 37 and 41 weeks based on this early scan were studied further. Fortnightly hospital antenatal visits were routine for most women from 28 weeks gestation.

A group of women who would be considered at low risk of giving birth to a SGA infant was identified by excluding the following: (a) women at presumed increased risk on the basis of their obstetric or medical history, for example a previous stillbirth or maternal hypertension, or socio-economic status and who were already identified as high-risk patients, (b) women who during the index pregnancy had had obstetric or medical problems such that they would be considered high risk, e.g. multiple pregnancy, antepartum haemorrhage, pregnancy-associated hypertension, and (c) those women in whom clinical growth retardation was suspected during the antenatal period and confirmed after delivery. In addition, women who attended for their first hospital antenatal visit after 12 weeks gestation were also excluded. In this way a minimum interval of 25 weeks between the first and last weight estimations was ensured since only women who were delivered between 37 and 41 weeks gestation, based on menstrual dates and ultrasound scan at between 16 and 18 weeks, were included in the analysis. Women with no record of weight at 28 and 32 weeks gestation, the time of the study period, were also excluded. Of the original 334 women 158 fulfilled the above criteria and they formed the study group.

The following characteristics of the patient, course of pregnancy and neonatal outcome were extracted from the case notes: maternal age, height, smoking habit, socio-economic status, parity, gestation and weight at booking, weight at 28 and 32 weeks gestation, overall weight gain, gestation at delivery, infant birthweight, and Apgar scores at 1 and 3 min.

Birthweight was adjusted to account for maternal height and weight and whether or not the infant was a firstborn and then compared with the birthweight standards of the Aberdeen survey by using the nomogram of Altman & Coles (1980). This defined a group of babies with birthweights below the 10th centile. The 158 women were therefore divided into two groups: those women who had babies with a birthweight below the 10th centile (SGA group) and those with babies of birthweight above the 10th centile, the appropriate-for-gestational-age (AGA) group.

Data were stored on a Vax 8730 minicomputer and analysed using the BMDP statistical software package (Dixon 1983). Differences between the groups were tested for significance with the χ^2 - and *t*-tests.

Results

All babies were liveborn vaginally. Of the 158 babies in the study group 30 weighed less than the 10th centile (SGA group) and 128 weighed more than the 10th centile (AGA group). There were no significant differences in Apgar scores between the two birthweight groups; in both groups the median score was 9 at 1 min ($\chi^2 = 0.02$, $P=0.88$) and 10 at 3 min ($\chi^2 = 0.20$, $P=0.65$). Table 1 details the maternal and pregnancy characteristics in the two groups. The only significant difference between the two groups was the maternal weight gain between 28 and 32 weeks gestation. In particular the mean booking weight and overall weight gain were similar for both groups of women and would be considered within normal limits for a low-risk antenatal population. A multiple logistic regression analysis showed that the effect of the weight gain between 28 and 32 weeks could not be explained by the slight differences in the other factors.

Of the 30 SGA babies 25 (83%, CI 69% to 97%) were born to women with a total weight gain of less than 1.4 kg over the 28–32-week period. The same cut-off in weight gain would have indicated a false-positive diagnosis in 42 of the 128 normal babies (33%, CI 25% to 41%).

Discussion

The increased risk to growth-retarded babies in terms of perinatal and infant morbidity and mortality is well known. Many factors are known to affect fetal size, some are associated with an increased risk of intrauterine growth retardation and are used to identify women who may be at high risk of giving birth to a growth-retarded baby. These include maternal height, parity and socio-economic status, maternal disease and smoking habit. Overall maternal weight gain in pregnancy is also associated with birthweight (Naeye 1979), but to our knowledge, the possible significance of poor weight gain over the period 28 to 32 weeks gestation in a group of women with a normal overall weight gain has not been recognized. It has been suggested that up to 70% of perinatal deaths associated with fetal

Table 1. Maternal and pregnancy characteristics in the two study groups

Characteristic	SGA group (n = 30)	AGA group (n = 128)	<i>t</i>	<i>P</i>
	mean (SD)	mean (SD)		
Maternal age (years)	26.7 (5.2)	26.4 (5.3)	0.3	0.77
Booking weight (kg)	61.2 (13.4)	58.6 (10.2)	1.2	0.23
Weight gain 28–32 weeks (kg)	0.99 (0.94)	1.95 (0.81)	5.61	<0.0005
Overall weight gain (kg)	12.58 (3.37)	12.83 (3.46)	0.37	0.71
Maternal height (cm)	164 (6.4)	162 (6.5)	2.15	0.31
	(n)	(n)	χ^2	<i>P</i>
Previous parity?				
No	10	54		
Yes	20	74	0.79	0.37
Smoker?				
No	19	92		
Yes	11	36	0.85	0.36
Socio-economic status				
I	0	10		
II	7	30		
III	12	51	6.5	0.16
IV	2	13		
V	4	5		
Other	5	19		
Gestation at booking (weeks)				
8	1	13		
9	5	13		
10	6	24	3.0	0.55
11	3	21		
12	15	57		
Gestation at delivery (weeks)				
37	2	4		
38	2	16		
39	9	24	5.1	0.4
40	8	45		
41	9	39		

SGA, Small-for-gestational-age, <10th centile birthweight; AGA, appropriate-for-gestational-age.

growth retardation could be prevented by identification of small-for-dates babies by 34 weeks gestation, which is the time when the increase in fetal body weight is at a maximum (Usher & McLean 1974; Chiswick 1985).

The antenatal diagnosis of fetal growth retardation is difficult and inaccurate; screening methods such as fundal height measurements or biochemical placental function tests (Hall *et al.* 1980; Barnard & Logan 1972) are insensitive in an overall antenatal population and have been superseded by ultrasonographic measurements. Neilson *et al.* (1984) developed a two-stage ultrasound schedule, with examinations in early pregnancy and at 34–36 weeks, and reported a 94% sensitivity and 90% specificity. The authors, however, failed to show any benefit

from its routine application in a low-risk population.

At the present time any clinical observation, especially a simple and inexpensive one, which might alert antenatal staff to the possibility of fetal growth retardation should be explored. Early detection of a potential case allows time to confirm the diagnosis by ultrasound and to plan management. The recording of maternal weight is a routine procedure and the recognition of a poor weight profile between 28 and 32 weeks gestation could confirm the diagnosis in more patients early enough to influence the outcome of the pregnancy. We are not claiming that poor maternal weight gain over this gestational period influences birthweight but rather that it may help to predict low birthweight. The population in

this retrospective study is unusual in that it only included patients who booked before 12 weeks, had a scan at 16 to 18 weeks, attended the antenatal clinic at 28 and 32 weeks gestation and were delivered between 37 and 41 weeks. This may explain the high proportion of women, 176 of the original 334, who had to be excluded from further study and why such a high proportion of babies, 19% (30 of 158), had a birthweight below the 10th centile. A number of low-risk women would be excluded simply because they were not reviewed at 28 and 32 weeks or because they did not have an ultrasound scan to confirm the gestation.

The birthweight nomogram of Altman and Coles, used to adjust birthweight for a number of factors, has been criticized (Carr-Hill & Pritchard 1983) and may not be applicable to the population in our hospital. Nevertheless, we felt some correction for the influence of maternal size, parity, gestation and sex of the infant had to be attempted.

Of the 30 SGA infants 83% had mothers who gained <1.4 kg between 28 and 32 weeks; so too had 33% of the normal infants. This specificity is poor, but in a low-risk population it compares favourably with a number of other variables which have been used to identify the problem (Villar & Belizan 1986).

In isolation, this observation would be likely to be of little value but might lead to better identification of SGA infants when used in conjunction with other measurements. It may also be appropriate to instigate more intensive antenatal care for those women whose weight gain seemed to be 'slowing down' during this time.

Static or minimal weight gain between 28 and 32 weeks did seem to predict a high proportion of SGA babies in this study, and if this observation is confirmed for other antenatal popula-

tions then further prospective evaluation is warranted.

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Serial oxytocin levels in amniotic fluid and maternal plasma during normal and induced labour

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Summary. Oxytocin was measured by a specific and sensitive radio-immunoassay in plasma and amniotic fluid after extraction with Sep-Pak cartridges in patients undergoing elective caesarean sections, normal labour and labour induced with oxytocin infusion or prostaglandins. In maternal plasma, levels of oxytocin correlated with the period of gestation; concentrations at term were significantly higher than in the first two trimesters. Maternal plasma levels of oxytocin before the onset of contractions were not significantly different from those at the onset of spontaneous labour or at full cervical dilatation. Levels of oxytocin in patients induced with oxytocin were not statistically different from levels observed in spontaneous labour. Amniotic fluid oxytocin levels were not significantly different between the groups either at the onset of labour or immediately before delivery. Umbilical arterio-venous differences in oxytocin were present in all groups except the prostaglandin-induced group. Increased prostaglandins associated with the onset of normal labour are probably not a consequence of changes in oxytocin concentrations.

In a previous report from this laboratory it was shown that oxytocin induction of labour did not lead to a marked increase in prostaglandins (PG) in the amniotic fluid despite successful initiation of uterine contractions (Padayachi *et al.* 1986; Reddi *et al.* 1984). This observation contrasts with those of Fuchs *et al.* (1981) who showed that oxytocin stimulates the production of PG *in vitro* from fetal membrane tissue. While oxytocin

from the maternal circulation can clearly induce uterine contractions independent of PG generation, the role of oxytocin from the fetal blood and amniotic fluid is unclear. The aim of this study was to investigate simultaneous concentrations of oxytocin in the umbilical circulation, amniotic fluid and maternal compartment to investigate further the precise role of this peptide hormone in the initiation of labour. In particular, we wished to look at the effect of infused oxytocin and intravaginal PG on amniotic fluid levels of this hormone.

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Patients and methods

The patients studied were multiparous African women from whom informed consent was obtained. Blood samples were obtained from 40 patients at between 7 and 44 weeks gestation. Thirty-seven patients at full-term gestation in labour or at the time of induction were divided into four groups.

Group 1. This group included eight patients delivered by elective caesarean section.

Group 2. This group included 13 patients in normal labour as assessed on the partogram (Philpott 1972).

Group 3. The third group included eight patients having labours induced with oxytocin. Synthetic oxytocin was infused at an initial rate of 2 mU/min, this was increased every 30 min until optimal uterine contractions were achieved; maximum rate of infusion was 32 mU/min. The membranes were ruptured at the start of the oxytocin infusion.

Group 4. The eight patients in this group had labour induced with prostaglandins. Prostaglandin E tablets (Prostin E, Upjohn) were inserted adjacent to the cervix. The dose was repeated every 2 h until labour began. Amniotomy was performed after the onset of regular uterine contractions and dilatation of the cervix. The patients in whom labour was induced were allocated in alternating sequence either to group 3 or to group 4.

The Bishop's score (Bishop 1964) was noted in groups 1, 3 and 4 and correlated with levels of oxytocin in the amniotic fluid and maternal circulation collected at the time of assessment.

Specimen collection

Amniotic fluid was collected hourly through an indwelling intrauterine catheter introduced vaginally, immediately after rupture of membranes in all patients for whom vaginal delivery was planned. In patients induced with PG, the first amniotic fluid sample was only obtained at the time of rupture of membranes. Amniocentesis was performed after exposure of the uterus in patients undergoing elective caesarean section.

Blood samples were collected at the same time as amniotic fluid into four evacuated tubes over 4 min from indwelling intravenous butterfly cannulas, because of the pulsatile release of oxytocin (Dawood *et al.* 1979; Otsuki *et al.* 1983a). Vaginal examinations were performed immediately after collection of samples to assess cervical dilatation. After the birth of the fetus and before the birth of the placenta the umbilical cord was clamped at two sites. Umbilical artery (UA) and umbilical vein (UV) blood samples

were obtained after which syntometrine was administered and the placenta was delivered.

The action of oxytocinase was inhibited by the collection of samples into pre-chilled EDTA tubes (Burd *et al.* 1985). These were transported on ice immediately to the laboratory where they were centrifuged at 4 °C at 2000 g and the plasma rapidly frozen in a dry ice/methanol bath. Phenanthroline, a recognized inhibitor of oxytocinase, was unsuitable in the radioimmunoassay, because it interfered with the antigen-antibody reaction at all concentrations. Under these conditions we demonstrated stability of oxytocin for at least 1 h. Plasma was extracted within 24 h onto Sep-Pak C₁₈ cartridges (Millipore, Waters Associates, Massachusetts), as described previously (Schams 1983) with only one modification in that elution from the cartridge was with methanol. Recoveries and extraction blanks were monitored in each batch. Recoveries were 106±3.2% and 108±4.2% at oxytocin concentrations of 40 and 100 µU respectively, and oxytocin was undetectable in the blanks.

Oxytocin assay

¹²⁵I-labelled oxytocin (specific activity of 2000 Ci mmol⁻¹) was purchased freshly labelled from Amersham (Bucks, UK). Specific antibody to oxytocin, used at a final solution of 1:112 000, was generously donated by Dr A. P. Flint (Babraham, Cambridge). Cross-reactions with lysine and arginine vasopressin were 0.46% and <0.003%, respectively (Sheldrick & Flint 1985). The assay was performed at 4 °C in a barbituric acid medium, pH 9.0 (Sheldrick & Flint 1981). Delayed addition of labelled oxytocin after 24 h pre-incubation of samples or standards with antibody resulted in a sensitivity of 10 µU/ml. All samples were assayed in duplicate.

Interassay coefficients of variance (CV) were 12.8%, 9.9% and 6.4% at 25, 50 and 100 µU/ml, respectively, whilst the intra-assay CV assessed at the same concentrations of oxytocin were 8.0%, 4.3% and 4.3%, respectively.

Statistics

Results were expressed as mean (SE) values. The significance of differences was assessed by the Wilcoxon signed rank test, Kruskal-Wallis analysis of variance and the Mann-Whitney

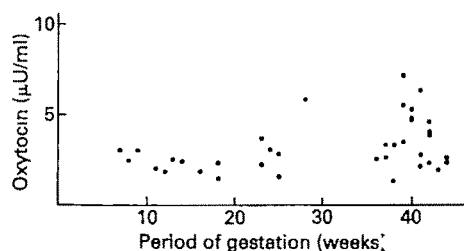


Fig. 1. Maternal plasma oxytocin correlated with period of gestation ($r = 0.39$, $P < 0.01$).

U-test where necessary. Results were deemed to be significant when $P \leq 0.05$.

Results

Oxytocin concentrations in maternal plasma correlated significantly with the period of gestation ($r = 0.39$, $P < 0.01$; Fig. 1). Oxytocin concentrations in the third trimester (mean 3.8, SE 0.33 $\mu\text{U/ml}$) were significantly higher than those in the first (mean 2.5, SE 0.2 $\mu\text{U/ml}$, $P < 0.05$) or second trimesters (mean 2.39, SE 0.24 $\mu\text{U/ml}$, $P < 0.02$).

Relevant clinical details of the patients in the four groups are presented in Table 1. No significant differences existed between the groups with respect to age, parity or gestational age at delivery.

Maternal oxytocin concentrations

The initial concentration of oxytocin was similar in the three groups of patients who were delivered vaginally, irrespective of whether they were having labour induced or were in early spontaneous labour (Table 2). During active labour, there was a significant increase in oxytocin only in those patients induced with an oxytocin infusion ($P = 0.02$; Fig. 2). Concentrations of oxytocin at full cervical dilatation were significantly higher in the patients induced with oxytocin than in those induced with PG ($P < 0.05$), but not statistically significantly different from

concentrations in patients in normal labour at corresponding stages of cervical dilatation.

Amniotic fluid oxytocin

Neither the initial nor the final amniotic fluid oxytocin concentrations differed between the groups (Table 2). No significant change with the progress of labour (Fig. 2) was present in any of the groups. Amniotic fluid oxytocin concentrations were significantly higher than the corresponding maternal plasma concentrations obtained at a cervical dilatation of 4 cm in all three groups ($P < 0.002$). At 8–10 cm dilatation, only oxytocin-induced patients and patients in normal labour demonstrated the same phenomenon ($P < 0.05$). There was a significant positive correlation between amniotic fluid and maternal plasma oxytocin levels collected simultaneously from patients receiving oxytocin infusion ($r = 0.45$, $P = 0.02$).

Umbilical blood oxytocin

There were no statistically significant differences in umbilical vein oxytocin concentrations between the four groups (Fig. 3) but umbilical artery oxytocin concentrations were significantly lower in the PG-induced group compared with the elective section group ($P < 0.05$) or the oxytocin-induced group ($P < 0.01$). There were no significant differences in umbilical blood oxytocin levels between patients who did not labour (group 1) and those who had spontaneous labours (group 2). Significant umbilical arterio-venous differences were present in babies born by elective caesarean section ($P < 0.02$), after spontaneous labour ($P < 0.05$) and oxytocin-induced labour ($P < 0.01$). In PG-induced labour, no difference was demonstrable.

In the group delivered by elective section there were significant correlations between umbilical artery and amniotic fluid oxytocin ($P < 0.05$, $r = 0.75$), umbilical artery and maternal venous oxytocin ($P < 0.02$, $r = 0.84$), umbili-

Table 1. Clinical characteristics of the four groups of patients

Patient group	n	Age (years)	Parity	Gestation (weeks)
1. Elective section	8	27.5 (2.2)	2.6 (0.5)	39.5 (0.4)
2. Normal labour	12	23.6 (1.3)	1.4 (0.2)	39.0 (0.3)
3. Oxytocin-induced	8	27.1 (1.1)	1.9 (0.4)	40.1 (0.9)
4. Prostaglandin-induced	8	27.6 (1.8)	1.9 (0.4)	41.1 (0.9)

Results are mean (SE) values.

Table 2. Amniotic fluid and plasma concentrations of oxytocin at amniotomy and at full dilatation in patients in induced and spontaneous labour

Patient group	Oxytocin concentration ($\mu\text{U/ml}$)			
	Amniotic fluid		Maternal plasma	
	Initial	Final	Initial	Final
1. Elective section	8.84 (1.38)			
2. Normal labour	8.04 (0.65)	7.77 (0.50)	4.05 (0.39)	5.48 (0.69)
3. Oxytocin-induced	8.40 (1.11)	9.54 (1.48)	3.55 (0.62)	6.07 (0.78)
4. Prostaglandin-induced	7.34 (1.27)	10.30 (5.05)	3.53 (0.47)	3.73 (1.13)

Results are mean (SE) values.

cal vein and maternal venous blood ($P < 0.02$, $r = 0.82$). Umbilical arterio-venous differences also correlated significantly with amniotic fluid oxytocin ($P < 0.05$, $r = 0.80$) and maternal venous oxytocin ($P < 0.05$, $r = 0.81$) in those women who did not labour. These correlations disappeared in the women who were delivered after labour.

The Bishop's score did not correlate with the initial amniotic fluid or maternal plasma oxytocin levels in the elective section or oxytocin-induced groups. A significant positive correla-

tion was present between Bishop's score and amniotic fluid oxytocin at amniotomy in the PG-induced group ($r = 0.74$, $P < 0.02$).

Discussion

This study has addressed two areas that have previously received little attention with respect to oxytocin and parturition. The first relates to amniotic fluid oxytocin levels during labour and to maternal and fetal oxytocin levels. The second is the influence of PG (particularly PGE)

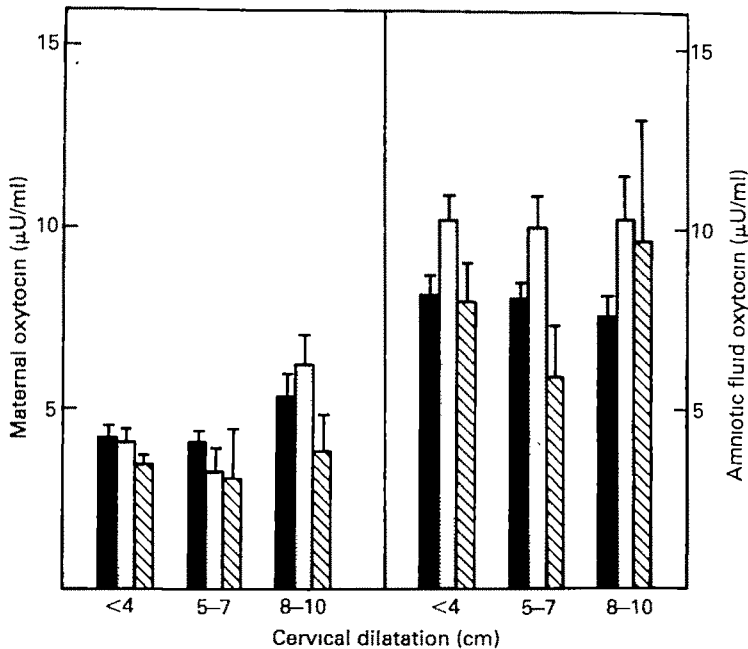


Fig. 2. Serial maternal plasma and amniotic fluid oxytocin concentrations at different cervical dilatations in patients in spontaneous labour (■), in oxytocin-induced labour (▨), and in prostaglandin-induced labour (▧). In oxytocin-induced labour plasma oxytocin was significantly higher at a cervical dilatation of 8–10 cm than at <4 cm dilatation ($P < 0.02$) and also significantly higher than in prostaglandin-induced labour at a dilatation of 8–10 cm ($P < 0.05$).

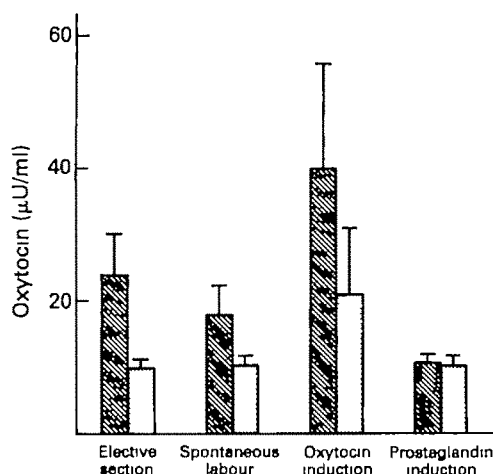


Fig. 3. Umbilical artery (▨) and umbilical vein (□) oxytocin concentrations in babies born after elective section, spontaneous labour, oxytocin-induced labour and prostaglandin-induced labour. Umbilical artery oxytocin concentrations were significantly lower in the prostaglandin-induced group compared with the elective section group ($P < 0.05$) or the oxytocin-induced group ($P < 0.01$).

on oxytocin levels in the materno-fetal unit. Both of these are important areas for the understanding of the role of oxytocin in the initiation and maintenance of human labour.

It is obvious from the data presented here that serial sampling of amniotic fluid throughout labour does not reveal any significant changes in oxytocin levels from early to late labour. Furthermore, induction of labour with oxytocin or PG does not influence the levels which are identical to those obtained before the onset of contractions. The fact that although maternal blood levels of oxytocin were higher in patients given intravenous oxytocin this did not affect amniotic fluid concentrations and therefore indicates that the predominant source in amniotic fluid is not from the maternal circulation. Fetal levels of oxytocin in the PG-induced group were low and yet amniotic fluid levels were equivalent to those observed in the other groups. This suggests that fetal levels also may not affect amniotic fluid concentrations to any significant degree. These observations are in contrast to the few limited studies reported previously. Dawood *et al.* (1978) claimed that amniotic fluid oxytocin levels at elective caesarean section were much lower than levels in late labour, while Seppälä *et al.* (1972) reported higher levels after labour. However, there were significant differences in

methodology between these studies and ours. Use of Sep-Pak cartridges led to much higher recoveries permitting detection of oxytocin in all the samples studied. Furthermore, samples of amniotic fluid were obtained regularly through an indwelling catheter throughout labour, scrupulously avoiding sampling periods associated with contractions or vaginal examinations. Other studies do not detail whether oxytocics were given before or after sampling of amniotic fluid; we avoided any possible influence from this source.

As a result, the present study has clearly established that there is no significant change in amniotic fluid oxytocin with the initiation of labour and its progress and a similar profile was found for maternal blood. Fuchs *et al.* (1983) also demonstrated no dramatic changes in plasma oxytocin in the first stage of normal labour. This raises the question of how oxytocin acts given the observation that no obvious changes in levels are found.

Fuchs *et al.* (1982) have claimed that decidua contains oxytocin receptors and that production of PG is under the influence of this peptide, possibly diffusing from the amniotic fluid or fetal blood. They have also demonstrated that levels of oxytocin receptors increase with gestation and peak during early labour. It is, therefore, possible that changes in receptor concentration which increase sensitivity to circulating oxytocin may be the mechanism by which labour is initiated (Fuchs *et al.* 1984). While the current study has not examined this possibility, we have previously shown that exogenous oxytocin administration does not materially influence amniotic fluid PG levels and, by implication, production by the decidua and membranes. There is, therefore, no obvious evidence for a changing sensitivity to oxytocin with respect to PG production at term.

Examination of the cord blood oxytocin levels confirmed the many previous observations of an arterio-venous difference in the peptide suggesting production by the fetus (Dawood *et al.* 1978; Sellers *et al.* 1981; Otsuki *et al.* 1983b). As shown previously (Dawood *et al.* 1978) umbilical vein levels increased with oxytocin administration but, surprisingly, the absolute levels in spontaneous labour were no different from those before labour started. Of particular interest is the fact that PG administration ablated the arterio-venous difference in oxytocin. This effect was primarily due to decreased levels in

the umbilical artery suggesting that PGE which crosses to the fetus reduces oxytocin secretion by the fetal hypothalamic pituitary axis or increases its metabolism by fetal tissues.

The precise dynamics of oxytocin between maternal and fetal compartments cannot be clearly defined by this study. There is strong evidence that the fetus produces oxytocin as demonstrated by the umbilical arterio-venous differences. Umbilical artery levels correlate closely with amniotic fluid levels either before labour ($r = 0.748$; $P < 0.05$) inversely following oxytocin induction ($r = 0.893$; $P < 0.01$), but not in spontaneous or PG-induced labour. The oxytocin concentration in the umbilical vein showed a poor correlation with maternal venous levels at the end of labour; however, levels were increased in association with oxytocin infusion. Overall, it does appear as if umbilical concentrations are altered by changes in the maternal compartment and, before labour, amniotic fluid concentrations are influenced by the fetal levels. There is some evidence at least to suggest that maternal factors such as increased oxytocin or PGE alter the fetal oxytocin concentrations but these appear to have little influence on amniotic fluid levels.

Whether increased oxytocin in the fetal compartment crosses to the maternal compartment in amounts sufficient to influence the uterus and membranes remains speculative.

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Postpartum pleural effusion

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Summary. A prospective ultrasound study investigated the reportedly high frequency of asymptomatic pleural effusions in the early postpartum period. The pleural space of 50 women within 1-45 h of delivery was scanned with a real-time sector scanner. Only one woman had a pleural effusion. This patient had severe pre-eclampsia with clinically apparent pulmonary oedema. Our findings do not substantiate the previously reported high frequency of pleural fluid in healthy women immediately after delivery.

Hughson *et al.* (1982) presented radiological evidence of a pleural effusion in a substantial number of apparently healthy women examined within 24 h after vaginal delivery. Having been alerted by the finding of bilateral pleural effusions in one such patient, they analysed, retrospectively, the chest radiographs of 112 women who, during 1977-1978, had been examined (for undisclosed reasons other than 'lack of antenatal care') immediately postpartum, and who, according to the medical records, had no significant cardiorespiratory disease. Close examination of the posterior-anterior and lateral radiographs revealed blunting of the costo-phrenic angle with thickening of the pleural stripe (which they term 'small effusion') in 42%, and a well-rounded meniscus extending up the chest wall ('moderate effusion') in a further 4%. They then conducted a prospective study of 30 postpartum patients and 30 healthy non-pregnant women aged between 15 and 40 years. They found a 'small effusion' in 63% of the study group and in 10% of the controls; one postpartum patient and none of the control subjects had a 'moderate effusion'. Ten of the postpartum patients identified as having an effusion agreed to have a lateral decubitus radiograph,

and the presence of free fluid was shown in seven of them. There was no noteworthy difference between those with and those without an effusion in respect of general medical history, pregnancy history, labour and delivery (including fluid intake).

Hughson *et al.* (1982) offered only tentative explanations of their findings, but concluded that, in the absence of relevant symptoms or of cardiovascular disease, the presence of a small pleural effusion observed postpartum is probably innocuous and should not prompt intervention.

We were intrigued by the reported findings and decided to carry out a similar investigation using ultrasonography. This has been shown to be superior to radiology in clarifying the solid or fluid nature of a pleural opacity, is more accurate especially where small effusions are present and permits the detection of amounts as small as 3-5 ml of pleural fluid (Doust *et al.* 1975; Grymiski *et al.* 1976; Lipscomb *et al.* 1981).

Patients and methods

The study included 50 postpartum patients (three of Indo-Pakistan origin, the remainder white Europeans) who agreed to have an ultrasound scan of their chest. Patient selection was entirely a matter of convenience, depending mainly on the time and day of delivery. The intervals between delivery and scan ranged between 1 and 45 h, but most (84%) were <24 h. Of the 50 women 29 were delivered vaginally

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Table 1. Mode of delivery and pathology in the 50 patients in the study

Mode of delivery	No. of patients	Noteworthy pathology
Spontaneous; no epidural	8	Two patients had a postpartum haemorrhage (500–800 ml)
Vaginal; epidural	20	One patient had a postpartum haemorrhage One patient had moderate pre-eclampsia
Forceps, epidural	1	Postpartum haemorrhage; evacuation of the uterus (general anaesthesia); scan
Emergency section; general anaesthesia; no epidural for labour	2	One patient had moderate pre-eclampsia
Emergency section; general anaesthesia; epidural for labour	2	One patient had moderate pre-eclampsia
Emergency section; epidural	1	
Elective section; general anaesthesia	4	One patient had SLE with lupus nephritis with superimposed moderate pre-eclampsia One patient had fulminating pre-eclampsia
Elective section; epidural	12	One patient had moderate pre-eclampsia One patient had fulminating pre-eclampsia

and 21 by section. The mode of delivery and relevant pathology of the patients are summarized in Table 1. The patients delivered vaginally or by emergency caesarean section under epidural analgesia received Hartmann's solution intravenously in quantity sufficient to sustain an acceptable level of blood pressure. Those undergoing elective caesarean section under epidural analgesia received, during 'pre-loading', intra-operatively and postoperatively (before scanning), either 1.5–4.0 litres Hartmann's solution or a smaller volume of Hartmann's solution plus 500 ml Haematcel.

An IGE RT 3000 real-time ultrasonic scanner with a 5-MHz electronic sector probe was used for all patients. Patients were scanned in the sitting position in bed or on a chair at an angle of 45° to the horizontal after they had been in this position for at least 5 min (to allow any pleural fluid present to collect in the costo-phrenic portions of the pleural space). Longitudinal views were then obtained of the diaphragm and costo-phrenic pleural space usually through liver or spleen in both parasternal, anterior and middle axillary lines. The patients were then sat forward at 80–90° to the horizontal and scans were carried out in posterior axillary and paraspinal lines bilaterally.

Results

In only one of the 50 patients was the presence of pleural effusion detected. She was a 37-year-old primigravida of Pakistani origin who was admit-

ted as an emergency with fulminating pre-eclampsia at 28 weeks gestation. A hurried elective section was conducted under epidural analgesia (400 ml human serum albumin and 1 litre Hartmann's solution being given for pre-loading of the circulation). The serum albumin was 22 g/l postoperatively and she was given further infusions of colloid and crystalloid solution. She developed clinical pulmonary oedema and received frusemide.

An ultrasound scan of the pleural space at 14 h after delivery showed a left pleural effusion (Fig. 1). The antero-posterior erect chest radiograph performed at the same time showed widespread interstitial pulmonary oedema but no costo-phrenic angle blunting (Fig. 2). The erect lateral chest film showed a posteriorly located left pleural effusion (Fig. 3).

The patient's general condition improved although her recovery was complicated by bronchopneumonia proven bacteriologically. Follow-up ultrasound scan on day 3 showed a resolving left pleural effusion with a similarly sized small right pleural effusion. Follow-up chest radiograph showed complete resolution of the pulmonary oedema and pleural fluid by day 9.

Discussion

Our results are somewhat different to those reported previously by Hughson *et al.* (1982) and Hessén (1951). In the present study the ultrasound scans of the chests were performed within

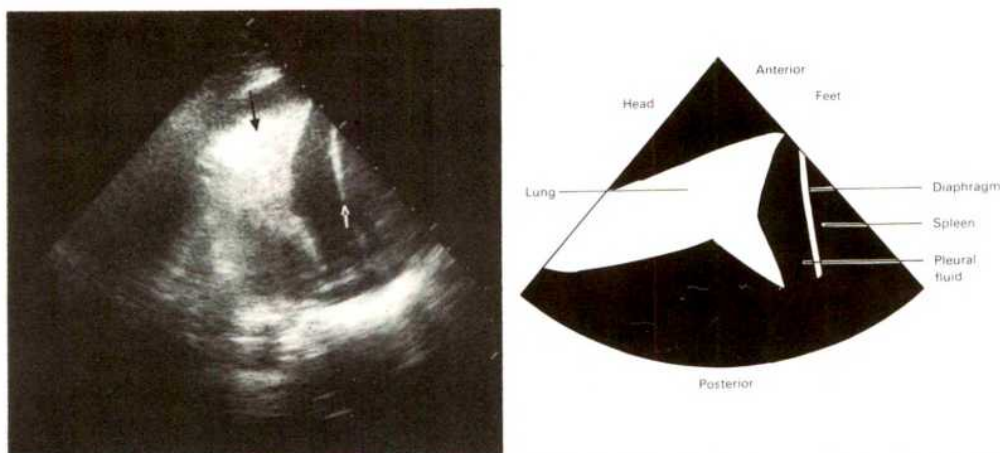


Fig. 1. Ultrasound scan and line diagram of the left pleural space showing densely reflecting diaphragm (white arrow) and lung (black arrow).

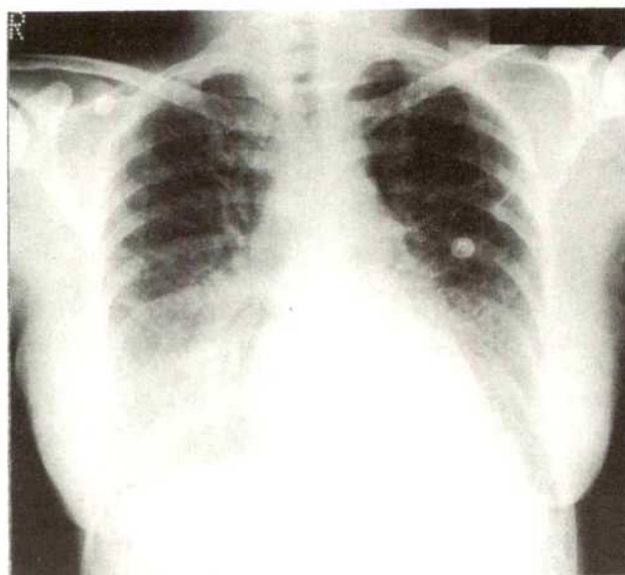


Fig. 2. Anterior-posterior erect chest radiograph (same patient as Fig. 1) showing interstitial pulmonary oedema.

24 h of delivery in 42 patients. (The remaining eight patients were scanned within 45 h of delivery.) Of these 50 patients only one had a pleural effusion and this patient was clinically in pulmonary oedema. None of our asymptomatic, clinically well patients had a pleural effusion.

It is well recognized that the posterior-anterior (PA) chest radiograph is unreliable in detecting small pleural effusions and 250–400 ml is the minimum amount that can be detected (Rigler 1931; Emerson 1924, 1928; Ganter

1923). Furthermore, blunting of the costo-phrenic angle on the PA radiograph is not a reproducible sign—it may occur with 175 ml of pleural fluid but as much as 525 ml could be present without blunting (Collins *et al.* 1972). Hughson *et al.* (1982) attempted to use lateral decubitus radiographs of the chest to confirm the presence of effusions they had diagnosed on PA films in the prospective part of their study. However, of the 10 patients who had lateral decubitus films only seven had the effusion confirmed.

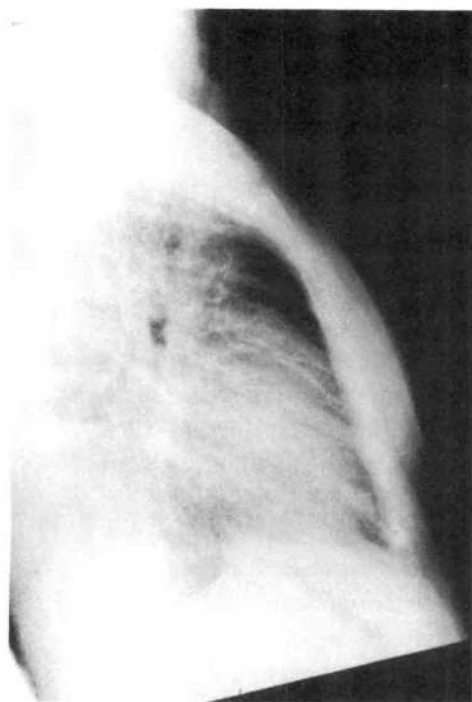


Fig. 3. Lateral erect chest radiograph showing posteriorly located pleural effusion (same patient as Figs 1 and 2).

Therefore, only 7 of 30 patients had a confirmed effusion even by radiographic criteria.

We used ultrasound scanning of the pleura to detect effusions as this has been shown to be reliable and sensitive. Doust *et al.* (1975) concluded that ultrasonography provides an accurate means of differentiating pleural fluid from other conditions which produce such opacities and is superior to the chest film as a means of localizing fluid. Grymiski *et al.* (1976) demonstrated the superiority of ultrasonic diagnosis of pleural effusion over radiologic techniques which included PA, lateral decubitus films and fluoroscopy where necessary. They

found that the difference was most marked in cases of small pleural effusion and that ultrasound permitted the detection of even 3–5 ml of loculated pleural fluid. Lipscomb *et al.* (1981) found ultrasound of the pleura superior to radiology in clarifying the solid or fluid nature of a pleural opacity.

We have found that pleural effusion does not commonly occur after delivery and believe that postpartum pleural effusions should be adequately investigated, explained, and appropriate management given, as in any other clinical situation.

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Uterine blood flow and myometrial activity at menstruation, and the action of vasopressin and a synthetic antagonist

ARNAR HAUKSSON, MATS ÅKERLUND, PER MELIN

Summary. Local endometrial blood flow was measured by a thermistor technique and myometrial activity by intrauterine pressure recording in 10 women before and during menstruation. The effect of lysine vasopressin infusion (1 pmol/kg body-weight per min) and of bolus injection of a synthetic oxytocin analogue, 1-deamino-2-D-Tyr(OEt)-4-Thr-8-Orn-oxytocin (10 nmol/kg body-weight), were studied. Spontaneous variations in blood flow were seen synchronous with clearly demarcated uterine contractions, the myometrial activity being significantly increased in early (day -1 to day +2) compared with late (day +3 to day +5) menstrual phase. The vasopressin infusion decreased blood flow, stimulated uterine activity and caused slight to moderate dysmenorrhoea-like pain. These effects were completely inhibited by the injection of the oxytocin analogue. In-vitro studies on uterine arteries confirmed that the analogue also inhibited the vasopressin-induced constriction of the uterine arteries. This antagonist was more effective than two other analogues, 1-deamino-2-D-Tyr(OEt)-4-Val-8-Orn-oxytocin and 1-deamino-2-Tyr(OEt)-oxytocin. The counteracting effect of 1-deamino-2-D-Tyr(OEt)-4-Thr-8-Orn-oxytocin on the vasopressin-induced decrease of blood flow and increase of contractions supports the therapeutic value of the drug in primary dysmenorrhoea and pre-term labour.

A variation in uterine activity at menstruation has previously been described (Bengtsson 1973; Lumsden & Baird 1985), but little is known about the uterine blood flow in women at this time of the menstrual cycle. Microtransducer

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catheters, allowing accurate recordings of myometrial activity, have been available for some time (Åkerlund *et al.* 1978), and lately, a thermistor technique for semiquantitative recording of local endometrial blood flow has also been developed (Hansson *et al.* 1987). In the present investigation we used these two techniques simultaneously for a study of uterine activity and blood flow in healthy women at menstruation.

It is believed that uterine ischaemia concomitant with myometrial hyperactivity plays an important causative role in the pain of primary dysmenorrhoea (Åkerlund *et al.* 1976). Vasopressin could be an important causative agent (Åkerlund *et al.* 1979) analogous to the suggested aetiological role of oxytocin in the exces-

sive uterine activity of preterm labour (Fuchs *et al.* 1982). Therefore, in the present investigation we also studied the effects of vasopressin and a synthetic antagonist, 1-deamino-2-D-Tyr(OEt)-4-Thr-8-Orn-oxytocin (Melin *et al.* 1986) on uterine blood flow and contractile activity. Apart from the in-vivo recordings of blood flow, an additional study of this and two other synthetic oxytocin analogues (Melin *et al.* 1986) on isolated uterine arteries was performed.

Patients and methods

Ten healthy women aged between 29 and 42 years (mean 36.3) with a parity of 1 to 3 and weight of 50 to 73 kg (mean 62.3 kg) were included in the investigation. They were well informed about the nature and purpose of the study and gave their written consent to participation. The project was approved by the local ethics committee.

Local endometrial blood flow was recorded by a thermodilution technique with two thermistors (YSI-520, Yellow Springs Instruments Company, Inc., Yellow Springs, Ohio, USA), one sensing the tissue temperature (thermometric thermistor) and the other, working at 5°C elevated temperature, sensing the heat loss caused by thermal conduction mainly due to the blood flow (anaemometric thermistor). The electrical power needed to keep the temperature difference constant between the thermometric and the anaemometric thermistors was recorded as a measure of flow. Myometrial activity was recorded as changes in intrauterine pressure by means of a microtransducer catheter with its sensor near the tip (PC-350, Millar Instruments, Houston, Texas, USA). The two thermistor catheters were attached to the pressure catheter. Details of these recording techniques have been given previously (Åkerlund *et al.* 1978; Hansson *et al.* 1987).

Recording of local endometrial blood flow and intrauterine pressure was performed in the period 1 day before up to 5 days after the onset of menstruation. Gynaecological investigation before the recording was judged as normal in all the women and blood pressure and pulse rate were normal.

Spontaneous variations in uterine blood flow and myometrial activity were recorded for at least 1 h. After this period, an intravenous continuous infusion of 1 pmol/kg body-weight/min of lysine vasopressin (Postacton, Ferring AB,

Malmö, Sweden) was given. After 40 min of vasopressin infusion, the women also received an intravenous bolus injection over 1 min of 10 nmol/kg body-weight of the oxytocin antagonist 1-deamino-2-D-Tyr(OEt)-4-Thr-8-Orn-oxytocin (Melin *et al.* 1986; Ferring AB). The vasopressin infusion and the recordings of uterine blood flow and pressure changes were continued for at least another hour after the injection.

Blood flow changes were measured by means of planimetry, in terms of the mean of added power per 10-min period for the total time recorded. Uterine contractile activity was quantified by planimetric analysis of the area under the recorded curve and expressed as the total pressure area per 10 min (Miller *et al.* 1976).

Statistical comparisons of blood flow changes in each of the subjects were performed by Wilcoxon's rank-sum test and of uterine activity by Student's *t*-test.

In-vitro studies

Segments of uterine arteries or their main branches were obtained from nine menstruating non-pregnant women undergoing hysterectomy for routine gynaecological indications. Longitudinal preparations with diameters of approximately 1.5 mm and lengths of 10 mm were mounted in tissue baths, and isometric tension was recorded on a Grass polygraph by means of force transducers (FT03). Basal tension was set at 0.5 g.

Arginine vasopressin was used at a concentration of 0.1 µmol/l and the inhibitory effect of 0.04–5.0 µmol/l of three analogues, 1-deamino-2-D-Tyr(OEt)-4-Thr-8-Orn-oxytocin, 1-deamino-2-D-Tyr(OEt)-4-Val-8-Orn-oxytocin and 1-deamino-2-D-Tyr(OEt)-oxytocin were tested (Melin *et al.* 1986). Inhibition with the antagonist was expressed as the response in percentage of the mean of two responses of the agonist when given alone and analysed as previously described for isolated myometrium (Melin *et al.* 1986).

The data were compared using Student's *t*-test.

Results

All the recordings were made with minimal discomfort to the participating subjects. No complications such as bleeding or subsequent infection occurred.

Spontaneous variations of blood flow, as indicated by the recordings of added power, were seen as transient reductions in phase with the uterine contraction, although occasionally increases were observed (Fig. 1 a & b). The most pronounced decreases were seen in women near the onset of menstruation (Fig. 1a) in whom the spontaneous contractions were most clearly demarcated and of highest amplitude. The added power varied between subjects, the mean values were 4.45 (SE 0.003) mW in the early and 4.39 (SE 0.05) mW in the late menstrual phase (range 4.20–4.54 mW) in the 10-min period before drug administration. The spontaneous uterine activity differed between early and late menstrual phases (Fig. 2), the total pressure area being significantly higher in the former ($P < 0.001$).

When infusion of lysine vasopressin was started the variations in blood flow in phase with uterine contractions diminished (Fig. 1 a & b). The blood flow decreased more rapidly in the late than in the early menstrual phase (Fig. 2), but at 10–20 min after the start of the infusion in the women in both phases a significant reduction ($P < 0.05$) was seen. The vasopressin infusion stimulated the uterine activity with increased frequency of contractions and elevated basal tone, the effect being maximal at approximately 15 min after start of the vasopressin infusion (Fig. 1a & b). The rise of total pressure area was statistically significant after 10–20 min of infusion ($P < 0.001$ irrespective of menstrual phase), the difference between the two groups persisting ($P < 0.001$; Fig. 2). Simultaneously with the changes in uterine blood flow and myometrial

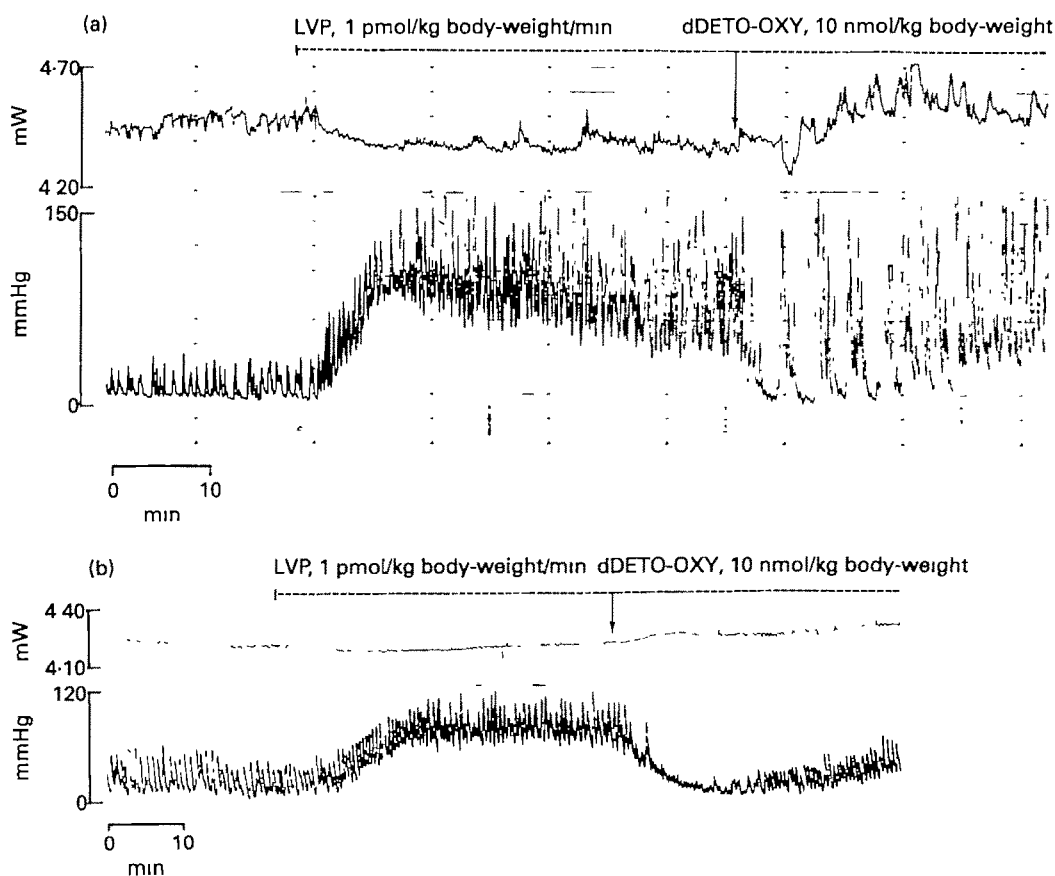


Fig. 1. Recording of uterine blood flow measured as added power (mW) and intrauterine pressure (mmHg) in non-pregnant women on day 1 (a) and day 5 (b) of menstruation. Infusion of lysine vasopressin (LVP) and bolus injection over 1 min of the vasopressin and oxytocin antagonist 1-deamino-2-D-Tyr(OEt)-4-Thr-8-Orn-oxytocin (dDETO-OXY) are indicated. The LVP infusion induced dysmenorrhoea-like pain in all subjects, which was completely relieved for about 15 min after antagonist injection.

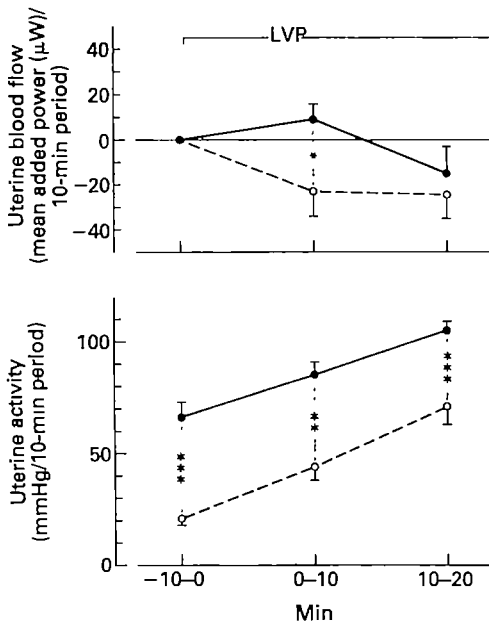


Fig. 2. The effect of intravenous infusion of lysine vasopressin (LVP, 1 pmol/kg body-weight/min) on integrated uterine blood flow (change from base line) and intrauterine pressure recordings (total pressure area) of women in early (●: day -1 to +2; $n = 6$) and late (○: day +3 to +5; $n = 4$) menstrual phase. Symbols represent means $\pm$ SE of results from each time period, and asterisks indicate significant differences (* = $P < 0.01$; ** = $P < 0.01$; *** = $P < 0.001$) between the two groups.

activity during vasopressin infusion all women experienced slight to moderate dysmenorrhoea-like pain.

The injection of 10 nmol/kg body-weight of 1-deamino-2-D-Tyr(OEt)-4-Thr-8-Orn-oxytocin during the vasopressin infusion caused increased blood flow and inhibited uterine activity (Fig. 1 a & b), with maximums and minimums in the second and first 10-min period after administration, respectively (Fig. 3). The increase of flow during the first and second 10-min period was statistically significant in both groups ($P < 0.005$), but there was a tendency for a more pronounced effect in the late menstrual phase (Fig. 3). The decrease of total pressure area after antagonist injection was statistically significant during 30 min after injection (Fig. 3; $P < 0.01$, $P < 0.001$ and $P < 0.01$ per 10-min period, respectively in both groups). Simultaneously with the increase of blood flow and decrease in uterine activity after administration

of the antagonist, the women reported relief of their vasopressin-induced dysmenorrhoea-like pain.

Analysis of variation in added power per 10-min period throughout the whole of the record showed a slight increase in basal flow ($P < 0.05$) in both early and late menstrual phase.

In vitro

Spontaneous contractile activity occurred in about half the vascular preparations. Arginine vasopressin (0.1 μ mol/l) invariably stimulated contractile activity and the analogues (0.04–5.0 μ mol/l) inhibited this effect in a dose-dependent way (Fig. 4). The most effective inhibitor was 1-deamino-2-D-Tyr(OEt)-4-Thr-8-Orn-oxytocin, followed in order by 1-deamino-2-D-

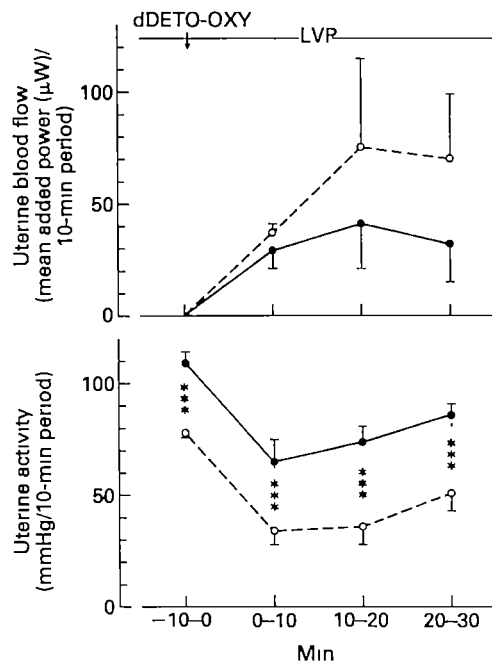


Fig. 3. The effect of intravenous injection of 1-deamino-2-D-Tyr(OEt)-4-Thr-8-Orn-oxytocin (dDETO-OXY, 10 nmol/kg body-weight) on integrated uterine blood flow (change from base line) and intrauterine pressure (total pressure area) recordings of women in early (●: day -1 to +2; $n = 6$) and late (○: day +3 to +5; $n = 4$) menstrual phase given intravenous infusion of lysine vasopressin (LVP, 1 pmol/kg body-weight/min). Symbols represent means $\pm$ SE of results from each time period, and asterisks indicate significant differences (*** = $P < 0.001$) between the two groups at each time period.

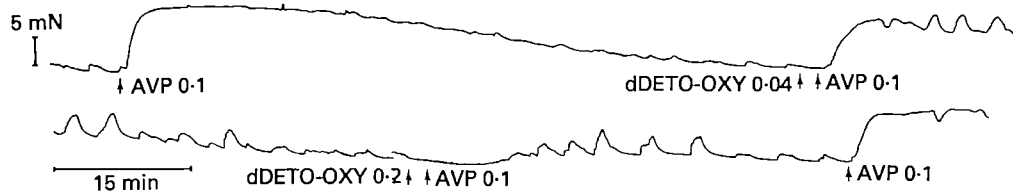


Fig. 4. Responses of an isolated human uterine artery preparation to arginine vasopressin (AVP) and inhibition with 1-deamino-2-D-Tyr (OEt)-4-Thr-8-Orn-oxytocin (dDETO-OXY). The concentrations of drugs are in $\mu\text{mol/l}$. The upper record continues below.

Tyr(OEt)-4-Val-8-Orn-oxytocin and 1-deamino-2-Tyr(OEt)-oxytocin (Fig. 5).

Discussion

In the present investigation the blood flow as recorded by thermoconduction technique did not differ significantly between the early and late menstrual phases. This agrees with the results of Prill (1959), using a thermocouple technique in recordings of endometrial blood flow. The inter-individual differences in added power found in the present study were presumably not caused by variations in the cyclical state of the endometrium, but by other factors, e.g. differences in the distance between probe and individual vessels.

In most instances well-demarcated uterine

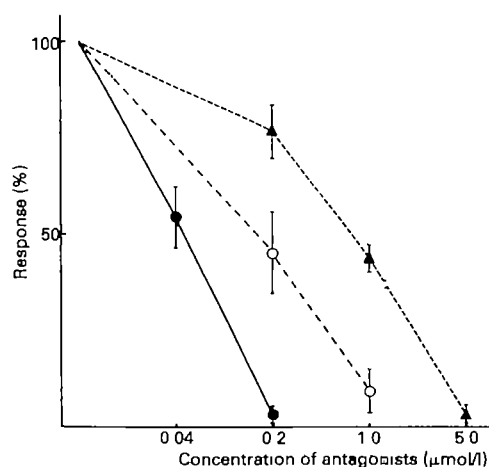


Fig. 5. Inhibition of arginine vasopressin (AVP, $0.1 \mu\text{mol/l}$) induced contractions of isolated, non-pregnant uterine artery segments by 1-deamino-2-D-Tyr(OEt)-4-Thr-8-Orn-oxytocin ($\bullet$ — $\bullet$), 1-deamino-2-D-Tyr(OEt)-4-Val-8-Orn-oxytocin ($\circ$ — $\circ$) and 1-deamino-2-Tyr(OEt)-oxytocin ($\blacktriangle$ — $\blacktriangle$). Preparations from nine non-pregnant women were obtained and the figures at each dose level of antagonist represent means $\pm$ SEM of results from 3 to 9 of them.

contractions caused a decrease in local endometrial blood flow, but occasionally transient flow increases were observed. A plausible reason for this finding is the fact that uterine contractions are often propagated (Martinez-Gaudio *et al.* 1973), and therefore redistribute blood flow from contracting to non-contracting areas, that is from areas of high resistance to areas of low resistance to blood flow. Another unexpected observation during the recordings before vasopressin and antagonist administration was the slight increase in basal flow over a period of hours. Since the present recording method does not have a drift in in-vitro models and is not influenced by variations in temperature (Hansson *et al.* 1987), this finding is likely to reflect a physiological phenomenon. A possible explanation could be that the presence of a probe in the endometrium for several hours could cause a reaction with hyperaemia. The long bed-rest (all subjects were supine throughout the recording sessions of 3–4 h) could also have contributed to the recorded change in flow. However, this increase in flow was slight and gradual and did not bias the quantification of the effects of uterine contractions or drugs given during the experiments.

The present study confirmed by means of quantification the previously described change of spontaneous uterine contractions during menstruation (Bengtsson 1973). Lumsden & Baird (1985), using the 'peak area', found a statistically significant decreased activity in dysmenorrhoeic, but not in healthy women, during the period of menstruation. However, those results are difficult to interpret in view of difficulties in defining basal tone, which is required for 'peak area' (Miller *et al.* 1976). The quantification method of total pressure area takes into account both individual contractions and the basal tone, and the present study demonstrated a significant difference in uterine activity between early and late menstrual phases. This change is probably the result of a varia-

tion in myometrial tissue concentration of ovarian hormones, which takes place with a time-lag to the decrease of plasma concentrations of oestradiol and progesterone before menstruation (Åkerlund *et al.* 1981).

In the in-vivo studies lysine vasopressin was used, since the human form arginine vasopressin is not a registered drug in Sweden but these two analogues are equipotent in their myometrial effects (Åkerlund *et al.* 1983). The vasopressin infusion appeared to have a slower influence on blood flow in early than in late menstrual phase. An explanation could be that in the early menstrual phase the persisting menstrual debris resulted in a less effective probe-tissue contact, which was then improved by the increased intra-uterine pressure caused by vasopressin, but spuriously giving a record of increased flow. This concept is supported by the lack of variation in the response to the antagonist, which was given after a firm probe-tissue contact had been established by the vasopressin-induced uterine hyperactivity.

Vasopressin infusion invariably stimulated the uterine contractile activity. In agreement with previous descriptions of cyclical variability in vasopressin effect (Åkerlund & Andersson 1976; Coutinho & Lopes 1968), there was a variation in absolute measures between early and late menstrual phases, but the responses were proportional to the degree of spontaneous activity.

The effects of vasopressin and the antagonist on the blood flow were caused not only by changes in uterine contractions, but also by effects directly on the smooth muscle of the arteries. This was suggested by the differences in times of maximum–minimum effects of the drugs *in vivo* and confirmed by the results of the in-vitro study. The latter showed that 1-deamino-2-D-Tyr(OEt)-4-Thr-8-Orn-oxytocin was the superior inhibitor of the three tested analogues and also that there was a five-fold difference in antagonistic potency between them. It is worth mentioning that the antagonism of the analogues on uterine arteries *in vitro* correlated closely with the potency these substances exhibited on myometrium from the non-pregnant human (Melin *et al.* 1986).

The counteracting effect of 1-deamino-2-D-Tyr(OEt)-4-Thr-8-Orn-oxytocin on the vasopressin-induced changes in blood flow and contractions, seen in the present study, could be the reason for the therapeutic effect of this drug in

primary dysmenorrhoea (Åkerlund 1987). An antagonistic effect of the drug on vasoconstriction and contractions caused by posterior pituitary hormones could also add to the beneficial effect of this oxytocin analogue in the treatment of preterm labour (Åkerlund *et al.* 1987).

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The influence of method of contraception and cigarette smoking on menstrual patterns

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Summary. Self-perceived menstrual patterns have been investigated in a sample of 2115 women aged 18-49 years using a postal questionnaire. Seven aspects of 'abnormal' menstruation were defined: prolonged periods, heavy periods, frequent periods, irregular periods, intermenstrual bleeding, painful periods and severe premenstrual syndrome. Women who used oral contraceptives were less likely than other women to report any of the seven menstrual abnormalities except for intermenstrual bleeding and severe premenstrual tension. Women who used an intrauterine contraceptive device (IUCD) reported prolonged, heavy, and frequent periods and intermenstrual bleeding more often than other women, but they did not report painful periods with undue frequency. Women who had had a tubal sterilization generally reported menstrual patterns similar to, or slightly less favourable than, women using no contraception or contraceptive methods other than the pill or the IUCD. There was a significant association between smoking habits and each of the abnormal menstrual patterns except for severe premenstrual tension. In every case, the effect of smoking was unfavourable and in almost every case, current smokers reported the worst experience, with ex-smokers occupying an intermediate position. We believe that these data are of considerable clinical significance and that they offer a basis for a conservative approach to managing menstrual disorders in some women.

There has been a rapid rise in the rate of hysterectomy in the UK during the past 30 years, while considerable differences exist in hysterectomy rates both within and between countries

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(Amirikia & Evans 1979, McPherson *et al.* 1981). A review of the literature failed to identify any temporal or geographical differences in levels of menstrual morbidity which might account for the observed variations in the rates of hysterectomy (Coulter & McPherson 1986). The review did, however, underline the increasingly discretionary nature of the decision-making process which results in a woman seeking, and a doctor providing, surgical treatment for disorders of menstruation. Accordingly, we decided to conduct a study to provide baseline epidemiological data about self-perceived menstrual patterns and the effect that certain socio-demographic factors have upon them.

Patients and methods

Of 2860 women aged 18 to 49 years, who were still experiencing menstruation and who were registered with two general practices in Milton Keynes, 2115 completed and returned a self-administered postal questionnaire. This represents a response rate of 74%. Questions were asked about menstrual patterns, general practitioner consultations for menstrual problems, and a number of socio-demographic characteristics including age, parity, social class, contraceptive practice and cigarette-smoking habit. The questionnaire had previously been validated by one of us (S.B.) in a pilot study which compared results obtained by post with results obtained by interview. We considered it important that the postal questionnaire should elicit replies as similar as possible to those which would be derived from a clinical consultation.

The results reported here are based upon each woman's subjective perception of her own menstrual pattern. It is well recognized that subjective and objective assessments of menstrual blood loss are poorly correlated (Stott *et al.* 1983). Despite this, we considered it relevant to investigate women's own perceived menstrual patterns as this is what is of importance to women themselves. In addition, a woman's perception of her own menstruation has a strong influence upon her decision whether or not to seek medical advice, and on the type of treatment prescribed by her doctors once advice has been sought.

Seven aspects of 'abnormal' menstruation during the year before the survey are considered in this report, based on the responses of the women to items in the questionnaire: (i) prolonged periods ('on average periods lasted 7 days or more'); (ii) heavy periods ('periods usually very heavy'); (iii) frequent periods ('interval between periods usually 21 days or less'); (iv) irregular periods ('interval between periods varied by 14 days or more'); (v) intermenstrual bleeding ('bleeding between main periods with most cycles'); (vi) painful periods ('pain experienced with most periods'); (vii) severe premenstrual syndrome ('very severely affected by symptoms in the week leading up to a period').

The associations between the seven abnormal menstrual patterns on the one hand and contraception and smoking on the other were analysed using two methods. First, the effects of

contraception and smoking were considered separately using a simple univariate method of analysis. Secondly, the effects of contraception and smoking were considered jointly using the GLIM statistical package. This multivariate approach also enabled us to take the confounding effects of age, parity and social class (variables which are not considered in detail in this report) into account.

Results

Table 1 summarizes the characteristics of the survey sample by age, parity, social class and cigarette-smoking habits. Contraceptive practices are shown in Table 2. Table 3 indicates that women who used an intrauterine contraceptive device (IUCD) were more often current cigarette smokers than women using other methods of birth control. This finding was unexpected in view of the frequently reported association between oral contraceptive use and cigarette smoking.

The associations between method of contraception and the seven abnormal menstrual patterns are shown in Table 4. Only the adjusted percentages are given because the figures do not differ much from those derived from the simple univariate analyses. As already indicated, the adjusted percentages were obtained using the GLIM statistical package, taking into account the effects of age (18–24, 25–34, 35–44, 45–49 years), parity (0, 1–2, 3+ children), social class (I–II, IIINM, IIIM, IV–V) and cigarette smoking (non-smoker, ex-smoker, current smoker). Simple additive logistic models were found to provide an adequate fit to each of the data sets. The numbers of women included in each analysis varied between 1573 and 1832 (i.e. between 74% and 87% of the total of 2115 women); this is because every data item had to be known to enable a woman to enter any given analysis.

Severe premenstrual syndrome was the only menstrual symptom not significantly associated with method of contraception. Women using oral contraception were less likely than other women to have reported any of the seven menstrual abnormalities except for intermenstrual bleeding and severe premenstrual tension. Women using an IUCD reported prolonged, heavy and frequent periods and intermenstrual bleeding more often than women in the other contraceptive groups. Unexpectedly, IUCD users did not report painful periods with undue

Table 1. Characteristics of 2115 women in the study sample*

	No. available for analysis*	% (n)
Age (years)	2096	
≤24		22.5 (472)
25-34		35.2 (738)
35-44		33.8 (708)
45-49		8.5 (178)
Parity (no. of living children)	1993	
0		30.1 (600)
1-2		52.5 (1046)
3-4		16.2 (323)
≥5		1.2 (24)
Social class†	1962	
I-II		35.4 (695)
IIINM		19.9 (390)
IIIM		31.0 (608)
IV-V		13.7 (269)
Cigarette smoking (no./day)	2092	
Never		48.1 (1006)
Ex-smoker		21.9 (458)
<15		18.1 (379)
≥15		11.9 (249)

*Women for whom a characteristic was unknown are omitted from the analysis.

†Registrar General's classification, based on husband's occupation if married and woman's occupation if unmarried.

Table 2. Method of contraception currently used by women in the study sample*

Method	%	(n)
Oral	27.4	(564)
None	18.6	(383)
Sheath	16.9	(348)
Vasectomy	14.2	(293)
Tubal sterilization	11.2	(231)
Intrauterine device	6.4	(132)
Withdrawal	2.0	(41)
Diaphragm	1.9	(39)
Rhythm	1.4	(29)

*Fifty-five women whose contraceptive practices were unknown omitted from Table.

frequency. Women who had had a tubal sterilization generally reported menstrual patterns similar to, or slightly less favourable than, women in the other/none contraceptive group.

Table 5 is similar to Table 4 except that it

considers the effects of cigarette smoking on abnormal menstrual patterns. As before, only adjusted figures are given and again simple additive logistic models described each data set satisfactorily. Contraceptive method (tubal sterilization, oral contraception, IUCD, other/none) was taken into account as well as age, parity and social class. Table 5 shows that there was a significant association between smoking habits and each of the abnormal menstrual patterns except for severe premenstrual tension. In every case, the effect of smoking was unfavourable, and in almost every case, current smokers reported the worst experience with ex-smokers occupying an intermediate position.

Discussion

The present study has a number of important limitations. First, it deals with self-perceived menstrual patterns reported by means of a postal questionnaire. These matters have already been considered in the methods section of the report. Secondly, although the overall response rate was 74% (a typical figure for postal surveys in our hands), missing values were fairly common and served to reduce further the effective sample size. Finally, the data are cross-sectional in nature. This complicates their interpretation in a number of ways. For example, women who in the past used an IUCD and who developed painful periods may have had the device removed. Again, women experiencing extremely severe menstrual symptoms may have had a hysterectomy and, as a result, have been excluded altogether from our analysis.

Bearing these caveats in mind, with the exception of intermenstrual bleeding and severe premenstrual tension, oral contraceptive users stand apart from other contraceptive users as having reported by far the fewest abnormal menstrual symptoms. This was the result we expected to find. The high prevalence of abnormal menstrual symptoms reported by IUCD users compared with women in the other contraceptive groups was also expected. Painful periods, however, were reported no more frequently by IUCD users than by women who had been sterilized or who were in the other/none contraceptive group. It seems unlikely that this finding could have arisen solely because women with painful periods had either avoided using or ceased to use an IUCD. Possibly, painful periods are not as much of a problem for IUCD users as

Table 3. Association between method of contraception and cigarette smoking (percentages with numbers of women in parentheses)*

Cigarette smoking	n	Method of contraception			
		Female tubal sterilization	Oral	Intrauterine device	Other/none
Never smoked	972	41.0 (93)	48.7 (273)	34.6 (45)	50.0 (561)
Ex-smoker	453	26.0 (59)	19.4 (109)	22.3 (29)	22.8 (256)
Current smoker	615	33.0 (75)	31.9 (179)	43.1 (56)	27.2 (305)

* Seventy-five women for whom contraceptive practice or smoking habit unknown omitted from Table.

Table 4. Association between method of contraception and the seven 'abnormal' menstrual patterns. The data shown are percentages of women with each pattern in each contraceptive group, adjusted for the effects of age, parity, social class and cigarette-smoking habits

Abnormal menstrual pattern	Number of women in analysis	Method of contraception				χ^2_{df} heterogeneity
		Tubal sterilization	Oral	Intra-uterine device	Other/none	
Prolonged periods	1777	14.2	5.3	22.7	13.3	49.6***
Heavy periods	1773	13.2	5.0	16.2	11.0	33.4***
Frequent periods	1734	3.3	1.8	6.6	2.1	7.8*
Irregular periods	1832	10.8	7.4	10.2	12.2	11.3**
Intermenstrual bleeding	1766	7.5	7.9	17.3	4.9	22.1***
Painful periods	1767	39.4	22.5	34.7	35.6	40.5***
Severe premenstrual tension	1573	4.3	4.4	4.0	5.0	0.5

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$

is commonly believed. The two symptoms which were particularly strongly associated with IUCD use were prolonged periods and intermenstrual bleeding. Although there is no evidence to suggest that intermenstrual bleeding associated with IUCD use is linked with any increased risk of malignancy, it would seem prudent to advise women to take account of the increased prevalence of these two menstrual symptoms when weighing up the benefits and risks of the various methods of contraception.

A frequently quoted British survey based on self-perceived menstrual patterns has suggested that menstrual blood loss increases subsequent to tubal sterilization (Neil *et al.* 1975). Another study which used an objective measure of menstrual blood loss found no such association (Kasonde & Bonnar 1976). These conflicting

reports raise the possibility that women who have been sterilized may hold a different view from other women of what constitutes an acceptable menstrual pattern. Women who choose to be sterilized may be a self-selected group who, before sterilization, have a lower threshold of tolerance towards menstrual symptoms than other women. Certainly, those who discontinue oral contraception to become sterilized are likely to be disappointed by their menstrual patterns after the operation. Again, it may be that once all prospect of future conception has gone, many women find menstruation less acceptable than before. Having said this, our study did not find much relation between tubal sterilization and abnormalities of menstrual patterns, although women who had been sterilized did rank second in severity within the four con-

Table 5. Association between cigarette smoking and the seven 'abnormal' menstrual patterns. The data shown are percentages of women with each pattern in each smoking group, adjusted for the effects of age, parity, social class and contraceptive method

Abnormal menstrual pattern	Number of women in analysis	Cigarette smoking			$\chi^2_{(2)}$ heterogeneity	$\chi^2_{(1)}$ linear trend
		Never	Ex-	Current		
Prolonged periods	1777	9.6	13.2	14.5	9.2**	8.6**
Heavy periods	1773	7.7	10.0	13.5	12.5**	12.4***
Frequent periods	1734	1.8	1.0	4.4	13.5**	8.8**
Irregular periods	1832	8.9	9.0	14.6	12.8**	10.7**
Intermenstrual bleeding	1766	5.4	6.3	9.2	7.8*	7.5**
Painful periods	1767	30.5	32.1	36.0	4.7	4.5*
Severe premenstrual tension	1573	4.4	4.5	5.3	0.7	0.6

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

traceptive groups for most of the menstrual symptoms studied. For painful periods they ranked first.

Associations between cigarette smoking and patterns of menstruation have not been so well documented in the literature as associations between contraceptive method and menstrual patterns. Several studies, however, have indicated that smokers experience more menstrual problems than non-smokers (Fries *et al.* 1974; Sloss & Frerichs 1983). In particular, smokers have been reported by Wood (1978) to experience more dysmenorrhoea and more prolonged periods than non-smokers, but these results did not control for the confounding effects of socio-demographic factors. Such effects have, of course, been taken into account in our study. Although it is possible that menstrual symptoms might lead to increased cigarette smoking (Barron 1984), recent work is pointing increasingly to a direct influence of the habit upon the endocrinological processes controlling menstruation (Greenberg *et al.* 1987).

We found cigarette smoking to exert an independent and important adverse influence on all menstrual symptoms studied, except for severe premenstrual syndrome. The fact that severe premenstrual syndrome was the one menstrual symptom which was not significantly associated with cigarette smoking lends further support in our view to the hypothesis that the influence of cigarette smoking upon menstrual patterns is a direct one. Thus it might have been expected that if women with a low threshold for menstrual complaint smoked more frequently than other women, they would have been particularly likely

to have reported severe premenstrual syndrome with high frequency.

The findings reported here have considerable clinical significance. They confirm and quantify the profound influence that different methods of contraception have upon menstrual patterns. They also provide further evidence about the harmful effects of smoking, and underline the need to reduce smoking in the population. We hope our results will be of use to general practitioners and family planning doctors when advising women with menstrual problems. Some believe that the present medical and surgical treatment of many menstrual disorders is too interventionist. Our data provide a basis for an alternative approach to managing menstrual disorders in some women.

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A comparison of gemeprost (Cervagem) pessaries and Lamichel tents for cervical preparation for abortion by dilatation and suction

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Summary. In a randomized, single-blind trial, 78 women having vaginal termination of pregnancy were treated with either a 1-mg gemeprost (Cervagem) pessary or a Lamichel tent inserted 3-4 h before operation. Both agents produced satisfactory cervical softening but dilatation was significantly easier after gemeprost. Pre-operative side-effects were more frequent after gemeprost but did not increase the need for analgesia. There were no differences between the two groups with regard to either postoperative symptoms and analgesic requirements or pre-operative and postoperative temperature, pulse rate and blood pressure. Gemeprost pessaries are easier, faster and less uncomfortable to administer.

The morbidity of abortion by dilatation and suction may be increased as a result of forcible dilatation of the uterine cervix and complications include immediate haemorrhage and, later, cervical incompetence and mid-trimester pregnancy loss. In addition, inadequate dilatation of the cervix makes complete evacuation of the uterus more difficult.

Pre-operative treatment to soften the cervix, making dilatation either easier or unnecessary, has been shown to be effective (Welch & Elder 1982; Chen & Elder 1983; Christensen *et al.* 1983; Christensen & Bygdeman 1984; Fisher & Taylor 1984; Welch *et al.* 1984). The two main types of pretreatment are the administration of prostaglandins (PG) as vaginal pessaries or the introduction of a cervical dilator. There has only been one report (Welch *et al.* 1984) of a comparison

between gemeprost (16,16-dimethyl-trans Δ_2 PG E_1 methyl ester, Cervagem) and Lamichel (a synthetic hydrophilic polymer impregnated with magnesium sulphate) and that was an open study where the operators were aware of the treatment given to each patient. We aimed to compare the use of gemeprost and Lamichel in a 'blind' study.

Patients and methods

Eighty patients admitted to Brighton General Hospital and Wistons Nursing Home, Brighton, for abortion by dilatation and suction at less than 16 weeks gestation agreed to participate in the study. It was initially intended to study 100 patients but for reasons of time the number was reduced. Gestation was determined from the last menstrual period confirmed by clinical assessment of uterine size. If there was a discrepancy ultrasound dating was performed. Patients were excluded from the study if they had active obstructive airway disease, cardiovascular insufficiency, elevated intra-ocular pressure, cervicitis or vaginitis or were currently receiving treatment for asthma. The study was approved by the district ethics committee. On admission, baseline temperature, pulse rate and blood pressure were recorded. Each patient was interviewed by one of

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us (N.D.) and asked to sign a consent form before being randomly allocated to receive either a 1-mg gemeprost pessary (Cervagem, May & Baker Ltd, Dagenham, Essex, UK) or a Lamitel tent (Cabot Medical Corp., Langhorne, Pennsylvania, USA) which were inserted by N.D. between 3 and 4 h before operation. Randomization was by numbered sealed envelopes containing the specific treatment allocation which were drawn in strict numerical order. Each batch of 10 envelopes contained five gemeprost and five Lamitel allocations randomly ordered.

To insert the Lamitel a Cuscoe's speculum was passed and the cervix and vault cleansed with antiseptic solution. A Lamitel tent, 5 mm in diameter, was inserted into the cervical canal using a sponge-holding forceps such that the tip was placed just distal to the internal cervical os. In cases where there was difficulty a 3-mm tent was inserted. In those allocated to receive gemeprost, a 1-mg pessary was inserted into the posterior fornix of the vagina. All patients lay on their beds for 1 h after treatment. Temperature, pulse rate and blood pressure were recorded by the nursing staff before premedication and also at other times if indicated. The patients were questioned about the occurrence of symptoms at these times and the details recorded. Both pre-operative and postoperative blood loss was assessed visually by the nursing staff and recorded as either none, minimal, moderate or heavy. The type of premedication given was standardized where possible but specific instructions were not included in the protocol.

In the operating theatre general anaesthesia was induced and the patients were placed in the lithotomy position. Information relating to the study was recorded on an individual case report form. With the surgeon outside the theatre, the assistant cleansed the perineum before passing a Sim's speculum into the vagina to remove either the Lamitel or all traces of the gemeprost pessary. The vagina was thoroughly cleansed with povidone iodine. The Lamitel was placed in a bag and removed from view. In this way, the surgeon was kept 'blind' to the treatment the patient had received.

When the assistant was ready, the surgeon (R.J.B. or C.W.H.) was admitted to the theatre from the 'scrub' room. A bimanual examination was performed and the bladder emptied if necessary. The time at which tenaculum forceps were applied to the cervix was recorded by the assistant as the start of the operation. Hegar dilators were

then passed sequentially starting with number 3 up to the size necessary to perform the operation. The force required to pass each dilator was assessed subjectively on a four-point scale—none, easy, moderate or difficult—and recorded on the case report form. The uterus was evacuated with a Karman suction curette and the products sieved through a bag suspended in the suction collection jar so that blood and fluid loss could be measured accurately. The time of the end of the operation was taken as the time of removal of the suction curette.

After leaving the operating theatre the patients were nursed in the recovery area before being returned to the ward. During the first 3 h after operation, temperature, pulse, blood pressure, blood loss, symptoms, complications and medication were recorded.

The results were analysed by the χ^2 -, Student's *t*- and Wilcoxon rank-sum tests as appropriate.

Results

Eighty patients were entered into the study. Two patients withdrew before receiving treatment, one changed her mind about having a termination and the other had a bad 'cold'. Two other patients were entered and received their allocation but were operated on an alternative operating list. No operative assessments were made but pre- and postoperative recordings have been included in the analysis. One other patient in the Lamitel group suffered a perforation of the uterus and required laparoscopy. Pre-operative readings and operative assessment of cervical dilatation only have been analysed. Overall, 38 patients received gemeprost and 40 Lamitel. On three occasions the 5-mm Lamitel could not be inserted and a 3-mm tent was used instead.

The two treatment groups were well matched with respect to age, parity, gestational age and previous utero-cervical surgery (Table 1). One patient in the gemeprost and two patients in the Lamitel group had had a previous vaginal induced abortion. Twenty-eight patients in the gemeprost group received temazepam, 10–20 mg, as premedication compared with 26 in the Lamitel group. In addition, two patients in the Lamitel group received intramuscular papaveretum. The remaining patients received no premedication.

The mean elapsed time between treatment and surgery was similar in both groups, 214 (SD 46) min for gemeprost and 226 (SD 40) min after

Lamigel ($t = 1.31$, $P = 0.19$). The mean duration of the operation was 5.1 (SD 4.1, range 2–21) min after gemeprost and 4.7 (SD 2.4, range 2–26) min after Lamigel ($t = 0.512$, $P = 0.61$) and there was no significant difference in the blood and fluid loss between the two groups, 123 (SD 175) ml after gemeprost and 152 (SD 197) ml after Lamigel ($t = 0.67$, $P = 0.5$). Table 2 shows the operative assessments. In addition to the small but statistically significant difference between the degree of dilatation at the start of the procedure there was a trend towards a greater percentage of cervixes being 'difficult' to dilate after Lamigel. At the two commonest dilatations for first trimester suction termination of pregnancy, Hegar 9 and 11, 26% and 43% respectively in the Lamigel group were described as 'difficult' to dilate for the first time compared with 16% and 23% in the gemeprost group.

After receiving gemeprost, patients experienced more unwanted side-effects before surgery (Table 3), notably abdominal discomfort and bleeding although discomfort was also experienced by half the patients receiving Lamigel.

Postoperative nausea, vomiting, abdominal

discomfort and vaginal bleeding were similar in both groups (Table 4). Analgesic requirements postoperatively were not significantly different. Nine of 38 in the gemeprost group and 9 out of 39 in the Lamigel group took oral analgesia whereas 4 and 2 respectively were prescribed intramuscular opiate analgesia.

Analysis of the degree of discomfort is tabulated in Table 5. Patients in the gemeprost group also developed more vaginal bleeding pre-operatively but there was no significant difference between the two groups with regard to pulse rate, blood pressure or temperature either pre-operatively or postoperatively. Two patients in the gemeprost group developed moderate bleeding and one a heavy loss associated with incomplete miscarriage.

Discussion

This is the first report of a blind, controlled study of the use of gemeprost (Cervagem) and a synthetic hydrophilic polymer impregnated with magnesium sulphate (Lamigel) before abortion by dilatation and suction. No control group was

Table 1. Patient details

	Gemeprost	Lamigel
Number of patients	38	40
Age (years) mean (SD)	20.9 (4.6)	22.0 (5.3)
range	(15–33)	(15–41)
Gestation (days) mean (SD)	76.5 (12.5)	73.2 (10.3)
Primigravida	35	36
Parous	2	2
Vaginal legal abortion	1	2*
Early miscarriage		1

*Including one of the two parous patients.

There were no significant differences between the two groups, χ^2 -test.

Table 2. Cervical dilatation in the two groups

	Gemeprost ($n = 37$)	Lamigel ($n = 39$)	<i>P</i> value
(a) Largest Hegar dilator inserted without meeting resistance median (range)	9 (5–13)	8 (5–11)	<0.001
(b) Largest Hegar dilator inserted to enable suction termination mean (SD)	10.6 (1.1)	10.4 (1.2)	> 0.2
range	(9–13)	(9–13)	
(c) Further dilatation required after noting resistance to dilator mean (SD)	1.8 (1.4)	2.9 (1.4)	< 0.001
range	(0–6)	(0–6)	

Analysed by Wilcoxon rank-sum test.

Table 3. Pre-operative side-effects in the two groups

	Gemeprost (n = 38)	Lamigel (n = 40)	P value
Nausea	3 (7.9)	1 (2.5)	>0.05
Vomiting	1 (2.6)	1 (2.5)	>0.05
Abdominal discomfort*	28 (74.0)	20 (50.0)	0.06
Vaginal bleeding	7 (18.0)	3 (7.5)	>0.05
None	9 (23.6)	20 (50.0)	0.03

*None required analgesia

Results are numbers of patients with percentages in parenthesis.

χ^2 -test.

Table 4. Postoperative side-effects in the two groups

	Gemeprost (n = 38)	Lamigel (n = 39)
Nausea	8 (21.0)	2 (5.1)
Vomiting	3 (7.9)	1 (2.5)
Abdominal discomfort	25 (65.7)	21 (53.8)
Vaginal bleeding	35 (92.1)	37 (94.8)
None	1	1

Results are numbers of patients with percentages in parenthesis.

There were no significant differences between the two groups, χ^2 -test.

Table 5. Analysis of abdominal discomfort

	Pre-operative		Postoperative	
	Gemeprost (n = 38)	Lamigel (n = 40)	Gemeprost (n = 38)	Lamigel (n = 39)
None	10 (25)	20 (50)	13 (34)	18 (46)
Slight to moderate	22 (53)	16 (40)	22 (58)	21 (54)
More severe	6 (15)	4 (10)	3* (8)	

*All transient.

Percentages in parentheses.

There were no significant differences between the two groups, χ^2 -test.

included as both treatments have previously been shown to be significantly better than placebo at softening the cervix and facilitating dilatation (Welch & Elder 1982; Chen & Elder 1983; Christensen & Bygdeman 1984; Fisher & Taylor 1984; Welch *et al.* 1984) and we considered that it would be unethical to withhold such treatment from any patient.

The number of surgeons was limited to two and unlike the only previous published comparison (Welch *et al.* 1984) they were 'blind' to the treatment received.

Laminaria tents have been shown to produce less satisfactory dilatation of the cervix compared with a 1-mg gemeprost vaginal pessary when

inserted 3 h before surgery (Christensen *et al.* 1983) but Welch *et al.* (1984) found no significant difference between the effectiveness of Lamigel 5 mm and 1 mg gemeprost given 2 and 3 h respectively before surgery. In this study we found that a significantly larger Hegar dilator could be passed without resistance in the gemeprost group and that significantly less further dilatation was required in this group after resistance to the dilator had been noted.

Although the unwanted effects of prostaglandins include nausea, vomiting, diarrhoea, abdominal cramps, vaginal bleeding and abortion, controlled studies of gemeprost *versus* placebo have shown no significant difference

between the amount of nausea, vomiting or diarrhoea (Chen & Elder 1983; Christensen & Bygdeman 1984; Fisher & Taylor 1984). The frequency of pre-operative abortion symptoms is related to duration of exposure to the prostaglandin and is minimal if gemeprost is given 3 h pre-operatively (Christensen *et al.* 1983).

Our finding of 28 out of 38 (74%) patients experiencing abdominal discomfort with gemeprost before operation is higher than previously reported (Chen & Elder 1983; Christensen & Bygdeman 1984; Fisher & Taylor 1984) although in 22 (79%) the discomfort was no more than moderate in degree and no patient required analgesia pre-operatively. In their comparison of Lamigel and gemeprost, Welch *et al.* (1984) reported mild, short-lived discomfort occurring soon after insertion of Lamigel in 50% (15 of 30) patients and this compared with our finding of discomfort in a similar proportion of patients but in whom it was not short-lived, being present at least until the time of premedication.

Apart from discomfort, Welch *et al.* (1984) reported no other side-effects before operation in association with Lamigel and they did not report the frequency of symptoms in their control group. Other trials of cervical pretreatment describe nausea, vomiting and diarrhoea in control groups (Chen & Elder 1983; Christensen & Bygdeman 1984; Fisher & Taylor 1984). In our study, one patient in the Lamigel group developed nausea and vomiting and three developed vaginal bleeding.

Our findings suggest that gemeprost 1 mg is slightly more effective than Lamigel at softening and dilating the cervix when both are given between 3 and 4 h pre-operatively. The prostaglandin analogue has more unwanted effects but there are problems with Lamigel. The insertion of the Lamigel tent requires the passage of a speculum into the vagina of an already anxious patient, the Lamigel can be awkward and uncomfortable to pass into the cervical canal and the process may need to be repeated with a smaller size; this was necessary in three of our patients. The procedure may take several minutes and involves the time of both a doctor and a nurse. Despite accurate placement, the Lamigel may fall out, as happened in 2 of 30 patients (Welch *et al.* 1984) and this might be discovered only at the start of the operation when it would

then not be possible to provide effective cervical softening.

Gemeprost pessaries have the considerable advantage that they are easy to insert and may be placed by a member of the nursing staff on the prescription of a doctor. As with all treatments patients receiving gemeprost should be fully informed of its intended purpose and the probability of some abdominal discomfort and bleeding. Analgesia should be readily available.

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Chlamydia trachomatis among sexually active teenage girls. Lack of correlation between chlamydial infection, history of the patient and clinical signs of infection

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Summary. Cultures for *Chlamydia trachomatis* were obtained from 1012 teenage girls attending an adolescence clinic mainly for family planning; *C. trachomatis* was isolated from 174 (17.2%). The proportion of chlamydia-positive girls varied between 15.7% and 28.5% depending whether or not there were symptoms and signs of infection. Neither the history nor the finding at pelvic examination offered conclusive evidence for or against the presence of an infection with *C. trachomatis*. Of the chlamydia-positive, untreated, asymptomatic girls, 17.5% developed symptoms of a genital infection within 3 months. There was a statistically significant difference in the proportion of positive cultures by the two investigators, suggesting that the procedure for specimen collection is of great importance.

Chlamydia trachomatis is among the most prevalent sexually transmitted pathogens and has been shown to be an important cause of genital infections (Thompson & Washington 1983; Taylor-Robinson & Thomas 1980). Women with a cervical infection are often asymptomatic and represent an important reservoir for the agent (Thompson & Washington 1983; Taylor-Robinson & Thomas 1980; Schachter *et al.* 1983). Since *C. trachomatis* has been linked with infertility it is especially important to investigate the prevalence of this agent among young, sexually active girls (Jones *et al.* 1982; Svensson *et al.* 1983; Weström & Mårdh 1983).

The aim of the present study, apart from

assessing the prevalence of *C. trachomatis* in a large group of teenage girls, was to investigate the relation between clinical signs, subjective symptoms, the history of the patient and chlamydial infection confirmed by culture.

Patients and methods

The study population consisted of 1012 girls attending an adolescence clinic for health care and family planning. During a period of 7 consecutive months in the years 1983-1984 all female patients whose visit required a pelvic examination were included. The mean age of the girls was 17 years (range 13-19).

The clinic is served by two nurse-midwives with training and experience in birth control and teenage counselling, who see around 3000 girls annually. The most common reason for attending the clinic is for contraception.

The girls were divided by chance between the two nurse-midwives and were interviewed before the examination. Information regarding age, purpose of the visit, current method of contraception, past history of sexually transmitted disease (STD) and pelvic inflammatory disease (PID) was obtained and recorded.

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In the present study 884 (87%) came for contraception. The remaining 128 (13%) came for urogenital symptoms or had a male partner with urethritis. Patients presenting with a current history of vaginal discharge, vulvar irritation, pruritus, dysuria, lower abdominal pain or dyspareunia were defined as having subjective urogenital symptoms.

A pelvic examination was performed and the girls were classified by the nurse-midwives as clinically infected or not, depending upon findings of purulent or mucopurulent endocervical exudate, erythema of the vaginal wall and/or oedema and fragility of any cervical ectopy.

Depending on their histories and the findings at pelvic examination the patients were divided into three groups: (A) Girls with clinical signs of infection and/or subjective urogenital symptoms and/or a male partner with urethritis. (B) Girls who were pregnant or had a past history of STD or PID, or both, but without current symptoms and signs of an infection. (C) Girls without a history of STD or PID and without current symptoms and signs of an infection. From groups A and B specimens for chlamydia culture were processed and the results received within 7 days. The culture-positive girls from these two groups received treatment within 10 days. Due to limited resources at the laboratory, specimens

from girls belonging to group C were frozen and kept at -70°C until cultured 8–10 weeks later.

The swabs were immersed in 2SP medium and kept at 4°C until they were transported to the laboratory within 24 h of the sampling. Cultures for *C. trachomatis* were made in cycloheximide-treated McCoy cells. All the cultures were read within 48 h except those from group C.

The specimens for chlamydia cultures were taken from the cervix with an ENT swab. No specimens were taken from the urethra.

The χ^2 analysis with Yates' continuity correction was used for statistical analysis.

Results

C. trachomatis was isolated from 174 (17.2%) of the 1012 girls. The distribution of positive cultures between the groups is shown in Table 1.

Group A. This group comprised 164 girls and 34 (20.7%) of them were chlamydia positive (Table 1). Of these 164 girls 128 attended the clinic because of symptoms or suspicion of an infection and in the other 36 girls the investigators had found signs of an infection. In 56 of the 128 girls with subjective symptoms the investigators also found signs of an infection and in this subgroup 16 (28.6%) were chlamydia positive (Table 2).

Group B. This group included 152 asymptomatic girls who were allocated to this group because of a past history of STD or PID (133 girls) or pregnancy (19 girls). *C. trachomatis* was isolated from 31 (20.4%) (Table 1).

Group C. This group included 696 girls. All specimens from this group were frozen and investigated 8–10 weeks after collection. A positive culture was obtained from 109 girls (15.7%; Table 1).

There was a statistically significant difference in the proportion of positive cultures obtained by the two investigators (Table 3). Nurse-midwife I, who invariably cleansed the portio before taking specimens for culture, obtained 20.1% positive cultures, whereas nurse-midwife II, who only cleansed when there was abundant discharge, obtained 14.3% positive cultures. Distribution of patients between the investigators occurred by chance. The difference in the proportion of positive cultures between the two investigators was especially pronounced in the group of girls where the samples were frozen.

Table 1. Distribution of positive chlamydia culture results in three groups of teenage girls

Patient group	n	Mean age (years)	Per cent chlamydia positive (n)	Mean age of positive girls
A	164	17.3	20.7 (34)	18.5
B	152	17.1	20.4 (31)	17.6
C	696	17.0	15.7 (109)	17.0

See text for description of groups.

Table 2. The distribution of subjective urogenital symptoms and clinical signs of infection and the number (n) and proportion of chlamydia-positive girls among girls belonging to group A

	n	Chlamydia-positive n (%)
Subjective symptoms only	72	10 (13.9)
Subjective symptoms and clinical signs	56	16 (28.6)
Clinical signs only	36	8 (22.2)

Table 3. The number of samples cultured at once or frozen and the proportion of chlamydia-positive cultures for each of the two midwives

	Midwife I		Midwife II	
	No. of samples <i>n</i>	Chlamydia-positive <i>n</i> (%)	No. of samples <i>n</i>	Chlamydia-positive <i>n</i> (%)
Non-frozen samples	143	32 (22.4)	173	33 (19.1)
Frozen samples	365	70 (19.1)	331	39 (11.8)
Total	508	102 (20.1)	504	72 (14.3)

The difference between the two midwives is statistically significant $P < 0.05$.

Follow-up of 109 chlamydia-positive girls from group C

Since the results from group C were not obtained until 8–10 weeks after the specimen collection, the 109 chlamydia culture-positive girls remained untreated during this period. Of the initially asymptomatic chlamydia-positive girls 103 were located and re-examined 10–12 weeks after the sampling. For the re-examination, the girls were again distributed by chance between the two investigators and not according to who had seen the girl at the previous examination.

During that period 18 (17.5%) had developed symptoms that suggested a genital infection and 12 of them had received antibiotic treatment elsewhere before the follow-up. Two had been admitted to hospital for suspected PID, one had been treated at an STD clinic because of partner's symptoms. The remaining nine had attended hospital clinics because of increased vaginal discharge in combination with lower abdominal pain and/or bleeding. Six girls had current symptoms and *C. trachomatis* was obtained from all six.

Of the remaining 85 girls, 68 were still asymptomatic and chlamydia culture positive, whereas 17 girls had a negative culture at the follow-up. Six girls could not be relocated and were lost to follow-up.

Discussion

C. trachomatis has been detected in the cervix from 4–33% of examined patients in different studies, depending on the study population (Thompson & Washington 1983; Taylor-Robinson & Thomas 1980; McCormack *et al.* 1979).

In the present study the proportion of chlamydia-positive girls varied between 15.7% and 28.5%, depending whether or not there were symptoms and signs of an infection. This isolation rate is high, especially in the asymptomatic group, but corresponds well to the prevalence reported from other adolescence clinics in Sweden (Örtqvist *et al.* 1984). The finding that positive cultures are only slightly more frequent among girls with symptoms and signs than among those without agrees with previous reports (Golden *et al.* 1984; Frazer *et al.* 1983).

The correct diagnosis of *C. trachomatis* by culture is subject to variations in sensitivity due to technical difficulties, including sampling, handling of specimens and culture procedures (Kallings & Mårdh 1982). The fact that the specimens from the asymptomatic group were frozen could have influenced the results, and would rather have decreased than increased the number of positive cultures. The study suggested that the sampling technique is important in general, and, in particular, when the samples were frozen. Since the results are based upon the number of infected cells in the samples, and since it is likely that freezing will damage a proportion of the cells, a careful sampling technique, aiming to recover as many infected cells as possible, is of importance for the sensitivity of the culture. Careful cleansing of the portio with a cotton swab before sampling seems to be one measure to improve the sensitivity.

Few reports have been able to demonstrate for how long an individual infected with *C. trachomatis* can remain an asymptomatic carrier. McCormack *et al.* (1979) reported that among 10 chlamydia-positive girls, four were still culture positive after 1 year. In the present study 67% of the untreated, asymptomatic girls remained chlamydia positive and asymptomatic after at least 10 weeks' observation. The study also demonstrated that almost one-fifth of the chlamydia-positive asymptomatic girls developed symptoms within 10 weeks of sampling. We can only suggest that their reported symptoms were actually caused by their chlamydia infection since they were seen and treated elsewhere and since there was no similar follow-up of the

initially chlamydia-negative girls to see if they had developed lower genital tract symptoms to any extent during the observation period.

One reason for the difficulty in establishing the possible duration of a silent carrier state is the clinical course of chlamydia PID, which is often characterized by few and mild symptoms and signs, in spite of which the infection often causes pronounced tissue destruction, sometimes leading to infertility (Weström & Mårdh 1983). Jones *et al.* (1982) found that approximately 10% of patients assessed for infertility with laparoscopic signs of tubal damage could not recall any previous episodes of genital infection. Therefore, the asymptomatic carriers of chlamydia constitute a risk group not only for further spreading of the infective agent but also for subsequent clinical infection and its sequelae.

An important and difficult issue in the study of sexually transmitted diseases is the sexual behaviour in the study group. Information regarding number of sexual partners and change of sexual partner was obtained. None of the girls in the asymptomatic chlamydia-positive group had changed sexual partners during the observation period, suggesting that they were silent carriers rather than reinfected.

All chlamydia-positive girls in the present study received treatment with tetracyclines, but no follow-up cultures were taken after treatment, in line with the routines of the clinic during the study period. Previous studies have shown that most of the patients are chlamydia negative when sampled 4–6 weeks after treatment (Lassus & Juvakoski 1982).

Young, sexually active girls belong to a high-risk group for *C. trachomatis*, which has been clearly demonstrated in the present study. It can also be concluded from the study that neither the history of the patient nor the finding at pelvic examination can offer conclusive evidence for or against the presence of an infection with *C. trachomatis*. It is recommended that young, sexually active girls be examined for *C. trachomatis* whenever they come to a clinic for contraception, that they be informed about the increased risk of infection when they change partners or have multiple partners and of possible means of protection against sexually transmitted diseases.

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Bacterial vaginosis: a double-blind randomized trial of the effect of treatment of the sexual partner

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Summary. In a double-blind randomized controlled trial we assessed the effect of metronidazole treatment of the male partner on the recurrence rate of bacterial vaginosis. Women who fulfilled the diagnostic criteria for bacterial vaginosis were treated with metronidazole given in single doses of 2 g on days 1 and 3. The sexual partners were randomized to receive either the same dosage of metronidazole or a placebo. A total of 107 pairs completed the study. One week after the start of treatment 89% of the women considered themselves improved or cured and 93% no longer had the diagnostic criteria for bacterial vaginosis. At assessment 5 weeks after the treatment, 75% reported that they were cured or improved and the diagnostic criteria were not present in 73%. Treatment of the male partner did not affect subjective symptoms, clinical signs and isolation rates of *Gardnerella vaginalis* at 1 and 5 weeks after treatment.

Bacterial vaginosis is now the recognized name of a clinical entity associated with colonization of the vagina with *Gardnerella vaginalis*. Bacterial vaginosis has been regarded as a sexually transmitted disease and treatment of the male partner is commonly advocated (Gardner & Dukes 1955; Dunkelberg 1977; Alawattegama *et al.* 1984). However, the effect of such treatment is largely unknown. In this randomized double-

blind clinical trial we studied the cure and recurrence rates in women treated with metronidazole after treatment of their sexual partners with metronidazole or placebo.

Patients and methods

Study population

We included in the trial 126 women, who attended a general practice or a gynaecological

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practice. They were eligible if they were non-pregnant and said they had one male partner aged 18 years or older. We did not include women who at the initial examination had candida or trichomonal infections detected by microscopy of vaginal secretions, or who had mucopurulent cervical discharge, tenderness of the uterus or adnexa or adnexal masses. The women had to satisfy at least three of the four following criteria for bacterial vaginosis (Amsel *et al.* 1983): (i) increased vaginal discharge, (ii) vaginal pH >4.5, (iii) detection of 'clue' cells in vaginal secretions at microscopy, and (iv) a positive amine test result.

The age and contraception use of the women who completed the study are shown in Table 1.

The women were informed of the purpose and the method of the study before they consented to participate. The partners received similar written information. The study was approved by the regional ethics committees.

Examination procedure

We examined the women before, and again 1 and 5 weeks after the start of treatment. At the gynaecological examination the lubricant on the speculum was not allowed to mix with the secretions collected for study. The pH was measured by pH-indicator paper in discharge pooled in the speculum or in the lateral vaginal fornix. We placed two drops of vaginal discharge on glass slides. One drop was mixed with isotonic saline and examined under a microscope. Trichomonads were identified as motile flagellated organisms. Clue cells were detected as vaginal epithelial cells with blurred cell borders. We added one drop of 10% potassium hydroxide to the other drop of vaginal secretion and examined for the release of a fishy odour. The slide

was examined with a microscope for the presence of hyphae.

We took samples for detection of *Chlamydia trachomatis* from the endocervix and the urethra with a cotton-tipped steel wire (Ent-swab) and this was placed in a sucrose phosphate buffer. Vaginal secretion for culture of *G. vaginalis* and *Trichomonas vaginalis* was obtained using sterile cotton-tipped charcoal-impregnated swabs and transported to the laboratory in Stuart's medium. We collected endocervical and urethral samples for culture of *Neisseria gonorrhoeae* at the first examination only.

Laboratory procedures

All specimens for culture were packed with a coolant and sent by mail. *C. trachomatis* was cultured in McCoy cells in vials treated with cyclohexamide as described by Ripa & Mårdh (1977). The swab for isolation of *G. vaginalis* and *T. vaginalis* was first streaked onto a chocolate agar plate, a 5% horse blood agar plate and a human blood agar plate. The two latter plates were also streaked with staphylococci for the detection of symbiotic growth. The plates were incubated in a 10% CO₂ atmosphere at 37°C for 24 h. We used the following criteria for a positive result: (i) slight symbiotic growth on 5% horse blood agar, (ii) haemolysis on human blood agar, (iii) pleomorphic Gram-variable rods and (iv) catalase-negative organisms.

After using the same swab to inoculate Diamond's medium, the secretion on the swab was mixed with a drop of saline and examined for trichomonads by direct microscopy. The cultures were examined after 3 and 6 days (Nielsen 1973). *Candida* species were identified by microscopy. Swabs for the isolation of *N. gonorrhoeae* were cultured as described previously (Lind 1975).

Table 1. Age and contraceptive method

Contraception	Treatment of male partner	
	Metronidazole	Placebo
Age of woman (years)		
Median (range)	29 (17-47)	28 (17-50)
Birth control pill	19	19
Intrauterine device	19	14
Barrier	11	4
None	5	16
Total	54	53

Treatment

The women were treated with metronidazole (Flagyl) tablets, 2 g on days 1 and 3. The partner was given the same treatment or a placebo. Eleven women infected with chlamydia and their partners were treated during the trial with oral erythromycin 500 mg twice a day for 10 days; eight pairs were given lymecycline 300 mg twice a day for 10 days.

Statistical methods

The partners were allocated to receive metronidazole or placebo by random allocation in blocks of four; the treatment received was unknown to the investigators until the completion of the study. The χ^2 -test was used and the results were considered significant at values of $P < 0.05$. We estimated the confidence limits (CI) for the difference between the two groups and the risk of type II error according to Feinstein (1975).

Results

We included 126 women in the trial, 63 partners were allocated to the metronidazole group and 63 to the placebo group. A total of 19 women were excluded: six from the metronidazole and four from the placebo group did not attend the second or third examination, three women and two partners did not take the tablets (two in the metronidazole and three in the placebo group), and three women (one in the metronidazole group and two in the placebo group) were treated again with metronidazole before the final analysis because their symptoms of bacterial vaginosis had recurred. One woman was excluded after detection of gonorrhoea at the first examination.

A total of 107 women and their partners completed the trial, 54 in the metronidazole and 53 in the placebo group. At the examination 5 weeks after treatment all women confirmed that they had had intercourse with their partner.

The diagnostic criteria for bacterial vaginosis could not be investigated at seven examinations because of menstruation. In a few examinations, measurements of pH, microscopy for clue cells and the result of the amine test were not reported (Table 3). The results of four cultures for *G. vaginalis* were lost.

We found no difference before or after treatment in symptoms, clinical signs, laboratory and culture results between the women whose partners were treated with metronidazole and those whose partners were treated with a placebo (Tables 2 & 3).

One week after treatment 95 of the 107 (89%) women treated with metronidazole were symptom-free or improved (Table 2). Five weeks after the treatment 80 of 107 (75%) still considered themselves cured or improved. Nine of the 12 women who were not better 1 week after

treatment considered themselves improved or were cured without further treatment 5 weeks after treatment, whereas 12/51 (24%) from the metronidazole and 12/44 (27%) from the placebo group had recurrent symptoms.

The difference between the two groups in the proportion of women symptom-free or improved at 5 weeks was not statistically significant (Table 2). The 95% CI of the difference was -14% to 19%. The risk of not detecting a 20% increase in subjective improvement in the metronidazole group was < 0.05 . If we include in the analysis the three women treated with metronidazole before the last examination, the subjective cure rates were 41/55 (75%) in the metronidazole and 39/55 (71%) in the placebo group; the difference between the two groups was not significant, the 95% CI was -13% to 20%.

The diagnostic criteria for bacterial vaginosis were fulfilled by 7/101 women (7%) after 1 week and by 28/105 (27%) after 5 weeks (Table 3). Five weeks after treatment the diagnostic criteria persisted in four of the seven women whose symptoms had persisted initially, one in the metronidazole and three in the placebo group. We detected recurrences in 12/50 (24%) of women whose partners were treated with metronidazole and in 12/48 (25%) of women with partners who received the placebo.

Clinical criteria of infection were absent after 5 weeks in 75% of the metronidazole and 71% in the placebo group (Table 3). The 95% CI of the difference was -13% to 21%. The risk of not detecting a 20% increase in the cure rate in the metronidazole group was < 0.05 .

We found *G. vaginalis* in samples from 93/105 (89%) of the women at the initial examination, in 27/106 (25%) after 1 week and in 35/106 (33%) after 5 weeks. At the 5-week examination *G. vaginalis* had disappeared from 13/27 (48%) of the women who still harboured the organism 1 week after the start of the metronidazole treatment. The organism reappeared in 8/41 (20%) of the women in the metronidazole and in 13/38 (34%) of those in the placebo group ($P > 0.05$). The symptoms reported after 1 and 5 weeks did not correlate with the presence of *G. vaginalis*.

We detected no difference in the subjective cure rates between women with and without *G. vaginalis* in the vaginal secretions at the first examination. Thus, 5 weeks after treatment 70/93 (75%) women who had *G. vaginalis* at the first examination reported improvement or com-

Table 2. The woman's symptoms according to the treatment of the male partner with metronidazole (M) or placebo (P)

	Time of examination					
	(1) Before treatment		(2) One week after treatment		(3) Five weeks after treatment	
	M (n = 54)	P (n = 53)	M (n = 54)	P (n = 53)	M (n = 54)	P (n = 53)
Vaginal symptoms						
Increased discharge	n 51 (%) (94)	48 (91)	20 (37)	15 (28)	23 (43)	20 (38)
Malodour	n 46 (%) (85)	41 (77)	6 (11)	3 (6)	12 (22)	12 (23)
Burning sensation	n 9 (%) (17)	7 (13)	4 (7)	0 (0)	1 (2)	4 (8)
Itching	n 22 (%) (41)	15 (28)	10 (19)	6 (11)	10 (19)	7 (13)
Improved or cured	n — (%)	—	51 (94)	44 (83)	41 (76)	39 (74)

Table 3. Diagnostic criteria for bacterial vaginosis in women according to treatment of the partner with metronidazole (M) or placebo (P)

	Time of examination					
	(1) Before treatment		(2) One week after treatment		(3) Five weeks after treatment	
	M (n = 54)	P (n = 53)	M (n = 54)	P (n = 53)	M (n = 54)	P (n = 53)
Clinical criteria						
Criteria for bacterial vaginosis present	n 54/54 (%) (100)	53/53 (100)	2/53 (4)	5/48 (10)	13/53 (25)	15/52 (29)
pH > 4.5	n 50/52 (%) (96)	49/52 (94)	19/54 (35)	17/51 (33)	24/52 (46)	21/50 (42)
Clue cells	n 47/53 (%) (89)	48/53 (91)	2/51 (4)	2/51 (4)	13/53 (25)	13/53 (25)
Positive amine test	n 50/54 (%) (93)	50/52 (96)	1/51 (2)	3/53 (6)	13/54 (24)	13/53 (25)
<i>Gardnerella vaginalis</i> detected	n 44/53 (%) (83)	49/52 (94)	13/54 (24)	14/52 (26)	14/54 (26)	21/52 (40)

Criteria for bacterial vaginosis: at least three of (i) increased, abnormal discharge, (ii) pH > 4.5, (iii) clue cells, (iv) positive amine test.

plete relief of symptoms compared with 8/12 (67%) of those without *G. vaginalis*. The criteria for bacterial vaginosis were met after 5 weeks by 24/91 (26%) of women with *G. vaginalis* at the first examination compared to 3/12 (25%) without *G. vaginalis*.

Five weeks after treatment, we found a pH > 4.5 in the vaginal secretions in 23 of 33 women (70%) who were positive for *G. vaginalis* compared with 22 of 68 (32%) who were negative

($P < 0.001$). Clue cells were present in 19/34 (56%) if *G. vaginalis* was isolated but only in 7/71 (10%) without *G. vaginalis* ($P < 0.001$). The amine test was positive in 20/35 (57%) in the presence of *G. vaginalis* compared with 5/71 (7%) in the absence of this bacterium ($P < 0.001$). The diagnostic clinical criteria were met by 20/31 (61%) with *G. vaginalis* in the secretions compared with 7/71 (10%) without the organism ($P < 0.001$).

The use of contraception did not affect the subjective or clinically diagnosed cure rates.

Trichomonas vaginalis was detected by culture in only one patient at the first examination and by microscopy 5 weeks after treatment in another patient. We found *Candida* species by culture in 17, 28 and 9 patients before and 1 and 5 weeks after treatment, respectively, but in only four of these patients at all three examinations. The presence or absence of *Candida* species had no influence on the results of the study.

Chlamydia trachomatis was isolated from 23 women (21%) at 38 examinations; 13 of the 23 infected women had one or two positive cultures at the first, 16 at the second and four at the third examination. Of the infected women 13 were from the metronidazole and 10 from the placebo group. We found *C. trachomatis* only in the urethral cultures in seven, in the cervix in 16 and in both sites in 15 women.

Treatment for chlamydial infection was given to one woman and her partner after the first examination, after the second to 18 couples, and to four couples after the third examination. We did not find *C. trachomatis* in any culture after treatment with erythromycin or tetracycline; however, we detected chlamydia in 14 of 18 women after metronidazole treatment but before specific antichlamydial treatment.

There was no difference in symptoms, signs and cure rates of bacterial vaginosis between women infected with chlamydia and those without this infection. Five weeks after treatment 15 of 23 (65%) women with chlamydial infection felt they were improved or cured compared with 65 of 84 (77%) of the women without chlamydia. The clinical criteria for bacterial vaginosis were still present 5 weeks after treatment in 7 of 23 (30%) women infected with chlamydia compared with 20 of 82 (24%) of those without this infection ($P>0.05$).

Treatment for chlamydia did not affect the cure rate of bacterial vaginosis. After 5 weeks 12 of 19 (63%) treated for chlamydial infection were subjectively improved or symptom free compared with 68 of 88 (77%) women who were not treated for chlamydial infection ($P>0.05$). The clinical diagnosis of bacterial vaginosis was made in 6 of 19 (32%) women who had been treated for chlamydial infection compared with 21 of 88 (24%) of those who had not received this treatment ($P>0.05$).

Discussion

Gardner & Dukes (1955) defined a condition they named '*Haemophilus vaginalis* vaginitis'. They considered the disease a specific infection caused by sexual transmission of the organism now called *G. vaginalis*. They detected the organism in 127 of 138 (92%) of women without other causes of vaginitis, but in none of 78 healthy controls.

McCormack *et al.* (1977) questioned this view as they found no association between colonization with *G. vaginalis* and an abnormal vaginal discharge. They isolated the organism from 29% of women who had no sexual experience and from 40% of women who had more than five partners. However, the study has been criticized because their information on sexual experience was based only on a confidential questionnaire.

In later studies *G. vaginalis* has been detected in 0–40% of asymptomatic women compared with 70–95% of women with a diagnosis of bacterial vaginosis (Pheifer *et al.* 1978; Amsel *et al.* 1983; Blackwell *et al.* 1983; Hill 1985). Osborne *et al.* (1982) isolated *G. vaginalis* significantly more often from women examined for symptoms suggestive of vaginitis than from asymptomatic controls.

In this study we detected *G. vaginalis* in 89% of 105 women who fulfilled the diagnostic criteria for bacterial vaginosis. The association of *G. vaginalis* with bacterial vaginosis was confirmed by our demonstration of a significant correlation between the isolation of the organism and the diagnostic criteria for vaginosis after treatment.

A diagnosis based solely on the isolation of *G. vaginalis* may be misleading as the symptoms and signs associated with bacterial vaginosis can be present in women without this organism. Anaerobic rods, including a group of motile curved rods named *Mobiluncus*, may be responsible for the symptoms (Holst *et al.* 1984; Hill 1985) and these organisms may coexist in symbiosis with *G. vaginalis* (Chen *et al.* 1979). Therefore, in this study, we based the diagnosis on the criteria proposed by Amsel *et al.* (1983) and they were also recommended by Eschenbach *et al.* (1984).

It was our intention to examine the effect of treatment after a diagnosis based only on office procedures. Consequently, we did not exclude women whose vaginal discharge was found on culture to contain pathogens other than *G.*

vaginalis, which could be responsible for discharge or vaginitis (with the exception of *N. gonorrhoeae*).

Metronidazole is significantly more effective than other antibiotics in the treatment of bacterial vaginosis (Pheifer *et al.* 1978; Balsdon *et al.* 1980; Malouf *et al.* 1981). Høvik (1983) compared different dosages of metronidazole and found that 2 g metronidazole given as a single dose on the first day and repeated on the third day was as effective as 400 mg given twice daily for 7 days in groups of 24 and 19 women, respectively.

In this trial we preferred the former regimen because of its simplicity. Our cure rate is similar to those reported by Høvik (1983) and Jones *et al.* (1985) who used the 7-day regimen.

Previous studies have not taken into account the possible influence of a concurrent infection with *C. trachomatis*. We found chlamydia in 21% of women with a diagnosis of bacterial vaginosis and without clinical evidence of cervicitis. This frequency is higher than the isolation rates of 5–10% commonly found in Scandinavian women (Mårdh *et al.* 1981).

Metronidazole had no effect on chlamydial infection as indicated by the re-isolation of chlamydia in 14 of 18 women after metronidazole treatment, but before treatment with specific antichlamydial drugs. In 11 of 19 women, treatment with erythromycin for chlamydia had no effect on bacterial vaginosis (Durfee *et al.* 1979) and tetracycline therapy in eight women was ineffective as treatment for bacterial vaginosis (Malouf *et al.* 1981). In our study the cure rate for bacterial vaginosis was similar in the women treated for chlamydial infection to that in the women not so treated. The high isolation rate of chlamydia in women with bacterial vaginosis would indicate that they may benefit from selective screening for this organism.

Gardner (1980) stated that most women whose sexual consort(s) remain untreated become reinfected. The concept of bacterial vaginosis as a sexually transmitted disease is supported by its rare occurrence in sexually inexperienced women. Further, *G. vaginalis* has been isolated from the urethra of 80–90% of partners of women with bacterial vaginosis, but not in male controls (Gardner & Dukes 1955; Pheifer *et al.* 1978).

After treatment for bacterial vaginosis, Pheifer *et al.* (1978) re-isolated *G. vaginalis* from

6 of 11 women who had intercourse with a new or untreated partner, compared with only 2 of 46 women without this exposure. They concluded, therefore, that such treatment may prove necessary to prevent reinfection.

Swedberg *et al.* (1985) treated 98 women for bacterial vaginosis with metronidazole in a single 2-g dose or with a 7-day regimen, and treated half of their male partners. They assessed 64 women at 21 days after treatment. Of the 39 women whose partners were treated 25 (64%) were cured compared with 17 of 25 (68%) of those whose male partners were not treated. However, the male partners were aware of the type of treatment, and cure rates were not based on the aforementioned diagnostic criteria, but on the disappearance of symptoms and on microbiological studies.

In our double-blind trial, the treatment of the male partner did not affect the cure rate of vaginitis after 1 and 5 weeks of treatment. By chance more pairs in the treated group used barrier methods for contraception (Table 1). If such methods protect against recurrence of bacterial vaginitis it would increase the likelihood of detecting a beneficial effect of metronidazole treatment. However, we found no difference in the cure rates according to the method of contraception, and no difference between the women who had treated and untreated partners. The number of women in the trial was so large, that it was probable that we would have detected a 20% increase in cure rate after metronidazole treatment of the partner. This speaks against sexual transmission of specific pathogens susceptible to metronidazole as the only cause of the disease.

We recommend that treatments which are used only by the female partner should be further investigated. The effect of local treatment with metronidazole has not been reported. Lactate gel (pH 3.5) therapy for 1 week in women with bacterial vaginosis has given immediate results comparable to the effect of 1-week treatment with metronidazole. However, the effect may be only symptomatic, and the recurrence rate after cessation of treatment was not reported (Andersh *et al.* 1986).

A possible explanation of the lack of treatment effects in the male consort could be a limited effect of metronidazole in the male owing to an absence of anaerobic bacteria. Trials of other treatments of the male partner, such as ampicillin, should be considered.

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Immunohistochemical study of cytoplasmic oestradiol receptor in normal, dysplastic and malignant cervical tissue

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Summary. Using a monoclonal antibody raised against an oestrogen receptor-related protein, p29, and an indirect immunoperoxidase technique to stain human tissue, the presence of the antigen was investigated in normal, dysplastic and malignant tissue of the uterine cervix. In normal tissue p29 was present throughout the ectocervix during the menstrual cycle and virtually absent from the endocervix. In dysplastic cervical tissue there was a decreasing p29 content with increasing severity of the dysplasia, and very low levels were seen in the carcinomatous tissues.

The cervix is a known oestrogen target in which the uptake and retention of oestradiol is due to the presence of specific, high-affinity binding proteins, the oestrogen receptors (Wiegerinch *et al.* 1980). Brush *et al.* (1967) reported that women injected with 50 μ Ci of tritiated oestradiol concentrated the isotope in the epithelia of their vagina and cervix. Sanborn *et al.* (1975) isolated and measured specific oestrogen receptors in the human cervix and, by analysis of tissue obtained by microdissection, found that

the concentration of oestrogen receptors was 5-10 times lower in the cervix than the endometrium.

Most investigations of oestrogen receptor structure and function have relied on the binding of radiolabelled compounds to the protein as the method of detecting the receptor. A consequence of this is that histochemical studies have largely relied on autoradiography, a difficult technique that necessitates long exposure times (Buell & Tremblay 1984).

Immunohistochemistry with antibodies against the receptor protein would circumvent these problems. During experiments designed to generate such reagents, a monoclonal antibody (D5) was identified that recognized a 29 kilodalton phosphoprotein (denoted p29) that was not itself the oestradiol receptor but was capable of interacting with the receptor (Coffer *et al.* 1985a). Further studies indicated that p29 was both qualitatively and quantitatively related to oestradiol receptor but not other steroid receptors or binding proteins (King & Coffer 1986). An immunohistochemical method has been developed for assaying p29 in human tissues (King *et al.* 1985). This method is capable of detecting p29 changes through the menstrual cycle in endometrium (M. Padwick, M. White-

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head and R. J. B. King, 1986 unpublished observations) and of predicting hormone sensitivity of human breast tumours (Cano *et al.* 1986). We have studied normal and neoplastic cervix with this method.

Materials and methods

Morphologically normal cervical tissue was obtained from 17 premenopausal patients who had a hysterectomy for benign gynaecological conditions not involving abnormalities of the cervix and who were not receiving hormone therapy of any description, apart from four who were taking combined oral contraceptive preparations. Colposcopic cervical biopsies were obtained from patients being investigated for cervical intraepithelial neoplasia (CIN), and colposcopic or punch biopsies were taken in patients with cervical carcinoma. A representative biopsy of cervical tissue was halved to provide one sample for routine fixation and paraffin embedding and one for immunohistochemical studies. The tissue for immunohistochemical study was fixed in 100% alcohol for 1 h (King *et al.* 1985) and then transferred to 70% alcohol solution before sectioning. Samples were then dehydrated, embedded in wax and cut into 6- μ m sections.

Monoclonal antibody

The monoclonal antibody D5 was generated from partially purified, soluble human myometrial oestradiol receptor (Coffer *et al.* 1977) and was purified from culture medium by ammonium sulphate precipitation, DEAE chromatography and protein A chromatography (Coffer *et al.* 1985a). It was stored at 4°C at 127 μ g/ml and diluted just before use. Under these conditions, no deterioration has been noted over a 1-year period.

Immunocytochemistry

An indirect immunoperoxidase method was used with alcohol-fixed, wax-embedded sections. Saline and control sections were taken through with each batch of specimen sections. The method has been validated elsewhere (King *et al.* 1985).

The tissue sections were dewaxed through two 2-min immersions in xylene, followed by 100% alcohol solution. The endogenous peroxidases

were then blocked by immersions for 15 min in a solution of 0.5% hydrogen peroxide in methanol. The sections were then placed in running tap water for 5 min and phosphate-buffered saline solution (PBS) for a further 5 min.

Dilute (1:4) sheep serum was added to the sections, 20 μ l per section, for 10 min. Excess serum was then carefully removed from each section and 25 μ l of the monoclonal antibody D5 (8 μ g protein/ml) was added to each section. The sections were left for 1 h in a humidified chamber at room temperature.

The sections were washed in two changes of PBS each for 10 min. From a solution containing 2 parts peroxidase-labelled, anti-mouse IgG (Amersham International, UK), 2 parts inert human serum and 47 parts PBS, 30 μ l of peroxidase-labelled antibody was then added to each section which was replaced in the humidified chamber for 45 min. After this, the sections were again washed in two changes of PBS for 10 min.

A solution of 1 mg diaminobenzidine hydrochloride, 5 ml of PBS and 5 μ l/100 vol.% hydrogen peroxide was then freshly made and 30 μ l added to each section for 10 min. The sections were washed in PBS for 5 min and distilled water for 5 min.

Finally, the sections were counterstained in haematoxylin for 5 s each before immersion in tap water for 5 min and then dehydration through 80% and 100% alcohol and xylene solutions for 2 min each before being mounted for microscopical examination.

Staining reaction

Positive staining for p29 was associated with a homogeneous golden-brown stain throughout the cytoplasm which varied in intensity with each tissue section.

The staining intensity of each tissue section was assessed and scored by one observer on a scale of 0 to 3.

Results

Normal cervical epithelium

Morphologically normal epithelium was studied in 17 patients (Table 1). Good cytoplasmic staining of the ectocervical squamous epithelium was obtained in all specimens (Fig. 1). There was no peroxidase reaction product seen within the

Table 1. Staining intensity (scored on a scale of 0 to 3) for an oestrogen receptor-related protein (p29) in ectocervix

Histology	No oral contraception/stage of cycle						Oral contraception used
	Proliferative		Secretory		Total		
	Mean (SD)	Median	Mean (SD)	Median	Mean (SD)	Mean (SD)	
Normal	2.5 (0.38)	3.0 (n = 8)	1.8 (0.49)	2.0 (n = 5)	2.4 (0.26) (n = 13)	2.3 (0.25)	2.5 (n = 4)
CIN I/II	2.5 (0.22)	2.5 (n = 6)	2.0 (0.58)	2.0 (n = 3)	2.3 (0.24) (n = 9)	2.6 (0.30)	3.0 (n = 7)
CIN III	1.8 (0.20)	2.0† (n = 5)	2.0 (0.22)	2.0 (n = 7)	1.9 (0.15) (n = 12)	2.1 (0.31)	2.0 (n = 11)
Squamous carcinoma	—		—		0.2 (0.12)* (n = 4)		

Significance of differences using Mann-Whitney *U*-test: between squamous carcinoma and other samples

* $P < 0.02$; between CIN III (proliferative) and normal (proliferative) † $P < 0.05$.

nucleus or associated with the cell membranes. There was homogeneity of staining throughout each section, the intensity score varying between 2 and 3 for all sections. In women not taking the combined contraceptive pill, the mean staining level was lower in the secretory than the proliferative phase of the cycle although the difference was not statistically significant. There was no difference between women using oral contraceptives and non-users (Table 1). The only staining observed in the ectocervical stroma was

in connection with smooth muscle in vessel walls.

There was very poor staining of any normal endocervical tissue examined with staining intensity scores of 0–1, although some staining intensity scores of 1 were noted in reserve cells as reported previously (King *et al.* 1985).

CIN III

Sixteen patients with CIN I/II were studied (Table 1). There was homogeneous cytoplasmic



Fig. 1. Normal transformation zone with squamous metaplasia. There is homogeneous cytoplasmic staining (intensity score 3) with D5 antibody in the squamous epithelium and an absence of underlying stromal stain. $\times 12.5$.

staining of the ectocervical tissue with no stromal take-up of antibody. The intensity of staining varied from 1 to 3. Neither the stage of menstrual cycle nor use of oral contraceptives influenced the staining pattern.

CIN III

There were 23 patients with colposcopically suggestive and histologically proven CIN III. The intensity of cytoplasmic staining varied from 1 to 3; most sections had a score of 2, both in the proliferative and secretory phases (Fig. 2). Neither the stage of menstrual cycle nor the use of oral contraceptives influenced the staining pattern.

Squamous carcinoma

Four patients presenting with stage I tumours were studied. Two tumours, histologically reported as poorly differentiated, exhibited no staining whatsoever with D5 (Fig. 3) although there was an adjacent area of CIN III in one section which had a staining intensity score of 1. The two other tumours were well differentiated with areas of keratinization. The staining in these sections was more heterogeneous, varying in areas from a staining intensity score of 0 to 2.

Adenocarcinoma

The one specimen of cervical adenocarcinoma we studied was negative.

Comparison of different types of ectocervix

The results are summarized in Table 1. The staining intensity scores were compared with the Mann-Whitney *U*-test. There were no statistically significant differences between any of the normal tissues and the CIN I/II tissue sections (Table 1). On the other hand, CIN III (proliferative) samples had significantly lower staining scores than either CIN I/II (proliferative) or normal (proliferative) samples (Table 1). These differences were not seen with either secretory phase samples or those from women using oral contraception.

Squamous carcinoma tissue sections stained less intensely than any of the normal or CIN tissue sections (Table 1).

Discussion

The monoclonal antibody D5 binds specifically to an oestrogen receptor related protein, p29. King *et al.* (1985) used D5 to investigate breast tumours and showed that the degree of peroxidase staining in the tissue correlated with

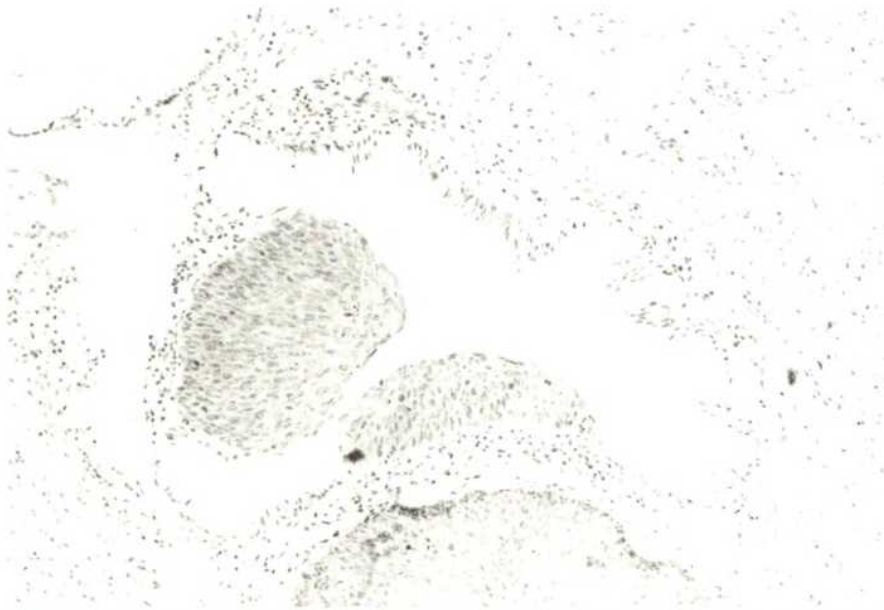


Fig. 2. CIN III extending into glands. Abnormal epithelium staining (intensity score 2) with D5 antibody. $\times 25$.

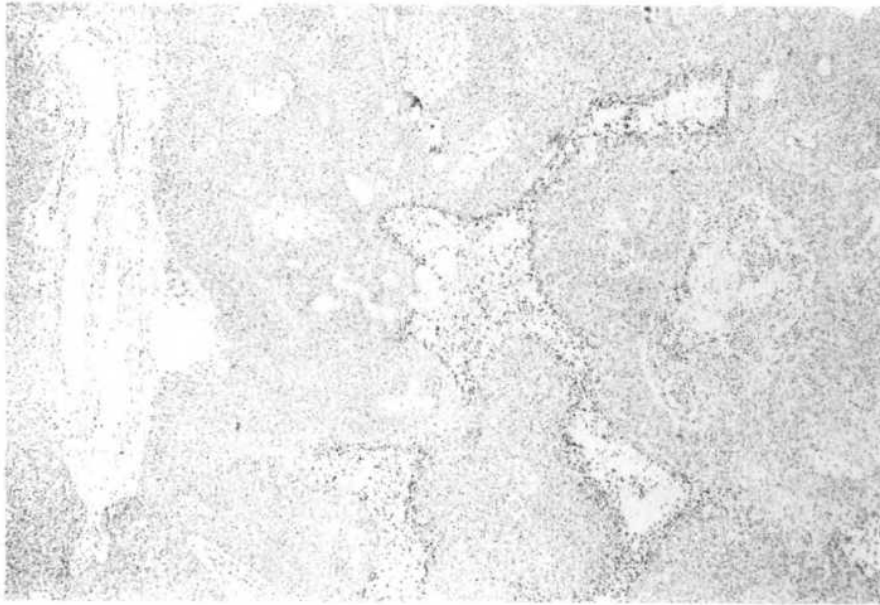


Fig. 3. Invasive squamous cell carcinoma. No staining after immunocytochemistry with D5 antibody. $\times 7.5$.

oestrogen receptor content. They also demonstrated that many structures in the female reproductive tract, including the cervix, showed positive staining with D5, and we believe that the amount of bound antibody, and thus the intensity of peroxidase staining, correlates with the amount of p29. It has also been shown that p29 content is qualitatively and quantitatively related to oestradiol receptor (King & Coffey 1986).

In recent years, several groups of workers have used immunohistochemistry with panels of monoclonal antibodies to investigate cervical wart virus infection, CIN and invasive disease (Morris *et al.* 1983; Fray *et al.* 1984). Overall, these studies have failed to show immunohistochemical discrimination between non-neoplastic and neoplastic cervical epithelia, although the findings of Morris *et al.* (1983) did provide support for the view that cellular proliferation of the type induced by the papillomavirus may represent an initiator stage in the process of CIN.

The purpose of the present study was to investigate immunohistochemically, using the monoclonal antibody D5, cervical cytoplasmic oestradiol receptors in normal and abnormal tissue and to explore any differences between neoplastic cells and their normal tissue counterparts. When peroxidase staining was present in

the tissues examined, it was contained entirely within the cytoplasm of the covering epithelium. There was no nuclear staining and no stromal staining. Analysis of the specimens of normal cervical tissue showed uniform staining intensity throughout the cytoplasm of the ectocervix throughout the menstrual cycle without any statistical difference in staining intensity between the proliferative and secretory phases. There was very little, if any, staining seen in the sections of normal endocervix. These findings contrast with those of Gould *et al.* (1983) who, using autoradiography to study nuclear oestrogen receptors, concluded that there was consistent stromal binding of receptors in both endo- and ectocervix but they concurred that there was little change in the nuclear receptor count within the squamous or columnar epithelia between proliferative and secretory phases of the cycle.

Specimens with histologically proven CIN again showed uniform cytoplasmic peroxidase staining of varying intensity. There was extremely limited staining of the sections of squamous carcinoma, especially in the poorly differentiated tumours. Although the overall number of specimens studied was small, the data are compatible with the idea of progressive loss of p29 from CIN I/II through CIN III to carcinoma.

In the one specimen of cervical adenocarcinoma, the complete absence of monoclonal staining was totally in keeping with similar findings in normal endocervix.

At present the selection of treatment for CIN and cervical carcinoma is based on clinical variables, in particular the stage of the disease and its suitability for local ablative treatment for dysplasia or radical pelvic surgery/radiotherapy for invasive disease. Neoplastic disease of the cervix is not considered to be hormone dependent, although Sugimori *et al.* (1976) did report a 5-year survival advantage in a group of patients with stage III disease treated by radiotherapy combined with oestrogen therapy. However, there appears to be no relation between receptor status and either stage of disease or the degree of differentiation of the tumour (Soutter & Leake 1987) and this is in keeping with the findings of Martin *et al.* (1986), that the survival curves for patients with oestrogen receptor positive and negative cervical tumours were virtually indistinguishable. It appears that any possible correlation between receptor content and prognosis will be independent of the present clinical and pathological indicators.

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Oral conception. Impregnation via the proximal gastrointestinal tract in a patient with an aplastic distal vagina. Case report

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Case report

The patient was a 15-year-old girl employed in a local bar. She was admitted to hospital after a knife fight involving her, a former lover and a new boyfriend. Who exactly stabbed whom was not quite clear but all three participants in the small war were admitted with knife injuries. The girl had some minor lacerations of the left hand and a single stab-wound in the upper abdomen. Under general anaesthesia, laparotomy was performed through an upper midline abdominal incision to reveal two holes in the stomach. These two wounds had resulted from the single stab-wound through the abdominal wall. The two defects were repaired in two layers. The stomach was noted empty at the time of surgery and no gastric contents were seen in the abdomen. Nevertheless, the abdominal cavity was lavaged with normal saline before closure. The condition of the patient improved rapidly following routine postoperative care and she was discharged home after 10 days. Precisely 278 days later the patient was admitted again to hospital with acute, intermittent abdominal pain. Abdominal examination revealed a term pregnancy with a cephalic fetal presentation. The uterus was contracting regularly and the fetal heart was heard. Inspection of the vulva showed no vagina, only a shallow skin dimple was present below the external urethral meatus and between the labia minora.

An emergency lower segment caesarean sec-

tion was performed under spinal anaesthesia and a live male infant weighing 2800 g was born, with Apgar scores of 7 and 9 at 1 and 5 min, respectively. On exploration, through the nearly completely dilated cervix, it was found that the normal uterus ended in a 2 cm deep vagina and that the vagina did not exist more distally.

The uterus, adnexa and renal tract appeared normal. Routine closure of the uterus and abdomen followed; a large tube drain was left in the uterine cavity.

While closing the abdominal wall, curiosity could not be contained any longer and the patient was interviewed with the help of a sympathetic nursing sister. The whole story did not become completely clear during that day but, with some subsequent inquiries, the whole saga emerged.

The patient was well aware of the fact that she had no vagina and she had started oral experiments after disappointing attempts at conventional intercourse.

Just before she was stabbed in the abdomen she had practised fellatio with her new boyfriend and was caught in the act by her former lover. The fight with knives ensued. She had never had a period and there was no trace of lochia after the caesarean section. She had been worried about the increase in her abdominal size but could not believe she was pregnant although it had crossed her mind more often as her girth increased and as people around her suggested that she was pregnant. She did recall several episodes of lower abdominal pain during the previous year. The young mother, her family, and the likely father adapted themselves rapidly to the new situation and some cattle changed hands to prove that there were no hard feelings.

The postoperative course was uneventful and the drain was removed on the 6th day. She started complaining about lower abdominal pain

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8 months postpartum while she was still breast-feeding. An attempt was made to construct a vagina by tunnelling between the urethra and rectum. A proximal vagina filled with old blood was found after 3–4 cm as described by Jeffcoate (1975). The tunnel was now dilated with appropriate instruments. It was found impossible to suture the skin of the introitus to the wall of the proximal vagina as advocated by Jeffcoate (1975) and criticized by others (Dewhurst 1981). A mould was constructed from a 20 cm³ plastic syringe cylinder with the distal end removed. The mould was fixed with nylon sutures to the labia and left in place for 2 weeks. The patient was discharged with a few oversized cervical dilators and instructed in their use. Follow-up was difficult, the patient was seen only some months later with a stenosed vagina and lower abdominal pain. Another reconstruction was attempted through scar tissue with much the same result.

Because she refused to have a hysterectomy and her cryptomenorrhoea was very painful, high doses of depot medroxyprogesterone were used to induce amenorrhoea. This was partly successful, but a hysterectomy became necessary to afford her relief from crippling pain when her son was 2.5 years old. By that time the son looked very much like the legal father.

Comments

A plausible explanation for this pregnancy is that spermatozoa gained access to the reproduc-

tive organs via the injured gastrointestinal tract. It is known that spermatozoa do not survive long in an environment with a low pH (Jeffcoate 1975), but it is also known that saliva has a high pH and that a starved person does not produce acid under normal circumstances (Bernards & Bouman 1976).

It is likely that the patient became pregnant with her first or nearly first ovulation otherwise one would expect that inspissated blood in the uterus and salpinges would have made fertilization difficult. The fact that the son resembled the father excludes an even more miraculous conception.

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Algodystrophy in pregnancy. Three case reports

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Case reports

Patient 1

A 25-year-old woman developed pain in her left leg extending from hip to foot in the sixth week of her second pregnancy. The pain was constant in nature, disturbed her sleep and was worse when weight bearing. She had experienced similar pain during her first pregnancy but at that time the pain alternated between both legs. These symptoms subsided spontaneously 6 months after the first pregnancy only to reappear during her second pregnancy. This time the symptoms were confined to the left leg. When first seen there had been no change in symptoms in the 10 months following her second pregnancy. On examination the left leg, from mid-thigh to foot, was cooler than the right. There was bony tenderness over the hemipelvis, greater trochanter, lower femur, tibia and patella on the left. Clinical examination and radiographs of the lumbar spine were normal and, in particular, there were no signs of a nerve root lesion. A bone scan showed marginally increased uptake of isotope in the left foot that was of doubtful significance. On the basis of pain associated with vasomotor change and diffuse bone tenderness a diagnosis of algodystrophy was made. Active physiotherapy resulted in resolution of her symptoms over 4 weeks.

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Patient 2

A 33-year-old primigravida developed pain in the right inguinal region at 26 weeks gestation. Two weeks later the pain had extended to both inguinal regions and the sacral area; she required crutches to walk. By 30 weeks she required hospital care because of pain and the limitation of her mobility. When seen at that time she had bony tenderness over her pelvis, sacrum and greater trochanters. There were no signs of a nerve root lesion. A plain film of her pelvis was normal. Epidural injection of 1% lignocaine completely abolished her pain for 24 h. For the rest of her pregnancy partial control of her pain was achieved with TNS (transcutaneous nerve stimulation). She had an uneventful vaginal delivery. After the birth, lumbar spine movements were normal. One week postpartum she could walk unaided and her pain resolved slowly over the ensuing 3 months. The nature of this patient's pain, the absence of a local inflammatory, neoplastic or neurological lesion and the diffuse bone tenderness strongly suggested that this patient had algodystrophy. The rapid response to physiotherapy after delivery would be consistent with such a diagnosis.

The physiotherapy in this case consisted principally of an exercise programme that promoted full weight-bearing without assistance. In most but not all patients such a programme is sufficient to relieve symptoms at an early stage.

Patient 3

A 29-year-old woman experienced mild low back pain during her second pregnancy. Her first pregnancy 3 years previously had been uneventful. After 1 h in the second stage of labour she complained of intense pain on the left side of the pelvis extending into the left thigh. The pain settled immediately after a forceps delivery. Ten weeks later she developed further low back pain

extending into the left thigh. The pain later extended into the left calf and foot. This pain was sometimes associated with parasthesiae. A diagnosis of sciatica due to a prolapsed intervertebral disc was made. She was managed accordingly but despite a wide range of treatments her symptoms worsened over the next 2 years. She was first seen in the department of rheumatology at this stage, at which time her symptoms were greatly restricting her exercise tolerance and she had difficulty coping with her two young children.

On examination there was a hyperaesthetic area over her low back extending over her left thigh. There was no indication of a nerve root lesion, in particular straight leg raising was normal and the tendon reflexes were normal. There was tenderness over the lumbar spine, pelvis and the left femur both proximally and distally. The whole of the left leg was cooler than the right. Peripheral pulses were normal. Radiographs of the hips, pelvis and lumbar spine were normal. A CT scan of her lumbar spine was normal. On the basis of pain, bone tenderness and vasomotor change, a diagnosis of algodystrophy was made. Active physiotherapy aggravated her symptoms. A series of sympathetic blocks (both epidural and regional) abolished the leg pain and reduced the severity of her back pain. Following these procedures the normal thermal symmetry in the lower limbs was restored. The patient still has some low back pain and tenderness over the sacrum.

Discussion

Algodystrophy is a syndrome in which pain is accompanied by autonomic dysfunction and trophic change (Evans 1947). Reflex sympathetic dystrophy syndrome and Sudeck's atrophy are synonymous terms. Transient osteoporosis of the hip (Hunder & Kelly 1968) and regional migratory osteolysis (Duncan *et al.* 1967) are related, if not the same condition, in which osteoporosis is the most prominent feature. The absence of well-defined diagnostic criteria in part relates to the wide spectrum of clinical features seen in this condition. The full clinical syndrome includes pain in a limb or part of a limb, dystrophic changes in the skin and its appendages, joint contractures, muscle wasting, hyperpathia, increased sweating and cyanosis, all involving the affected limb. Osteoporosis is the only radiological sign. Bone scintigraphy

may show increased isotope uptake (Kozin *et al.* 1976), though normal and decreased (Majd 1979) uptake have also been reported. In the presence of all these features there is little diagnostic difficulty, but difficulty arises when few of these signs are present or when they are present only in a mild degree. In such instances the two essential features are pain and evidence of autonomic dysfunction usually seen as alterations in the limb temperature which may be labile or constant (Coughlan *et al.* 1987).

Trauma, either accidental or surgical, is the most commonly cited precipitating event. Metabolic disease such as diabetes (Lequesne 1970), thyroid disease (Duncan 1928) and hyperlipidaemia (Pinals & Jabbs 1972) and drugs, isoniazid (Good *et al.* 1965) and phenobarbitone (Van de Korst *et al.* 1966), may also precipitate algodystrophy. Algodystrophy and transient osteoporosis of the hip have been reported in relation to pregnancy (Curtis & Kincaid 1959). It is unclear whether this association is due to a metabolic change in pregnancy or to trauma associated with the gravid uterus or delivery.

The four episodes of algodystrophy in our three patients were all associated with pregnancy and illustrate a number of points about algodystrophy. All three patients had pain that was not due to a specific mechanical, radicular, inflammatory or neoplastic lesion. In the two patients with unilateral and peripheral involvement, the diagnosis was facilitated by the finding of local cooling in the affected limb. All three had bony tenderness not related to a specific joint or soft tissue structure. The patient with bilateral hip involvement was a greater diagnostic problem. The extensive pelvic tenderness suggested that she did not have a local articular lesion. The response to epidural injection of lignocaine beyond the time of its expected therapeutic effect is a feature commonly seen in algodystrophy. If mild osteoporosis was present the bilateral nature of the lesion precluded its detection by comparing a normal and affected side. In the other two patients osteoporosis was not present, a bone scan in the first patient showed minor asymmetry.

Having established the diagnosis, all three responded quite quickly to three different accepted forms of treatment but in the second patient response only started after delivery. Our third patient still has residual symptoms. If one waits for the full range of clinical features to develop there will be an unnecessarily long delay

in initiating therapy. Indeed two of the patients had symptoms for over 6 months without developing the changes of severe or advanced disease. Many authors have emphasized the importance of early treatment in obtaining a good outcome. There is no one treatment that is effective in all cases of algodystrophy. Active mobilization with physiotherapy is often sufficient to reverse the process if undertaken at an early stage. Such a regimen may not be effective in some patients, particularly those with severe or chronic disease. In these circumstances when physiotherapy has not resulted in improvement or, as in our third patient, causes an exacerbation of pain, then other options include sympathetic blockade, high-dose corticosteroids and calcitonin. The use of beta-blockers, nifedipine and griseofulvin has been reported to benefit some of these patients.

Algodystrophy should be considered as a possible cause of pelvic or lower limb pain that occurs during or soon after pregnancy. The most informative features are local cooling of the affected limb due to autonomic dysfunction and bone tenderness that is not localized to an anatomic structure. As in these patients, local inflammatory neurological and mechanical lesions need to be excluded by appropriate investigations where indicated.

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Hereditary angioneurotic oedema and pregnancy. Case reports and review of the literature

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Patient 1

A 27-year-old primigravida with a history of hereditary angioneurotic oedema (HAE) since the age of 7 years had been well controlled on stanozolol. Before pregnancy she suffered episodes of swelling of the hands, face and abdominal pain. When her face swelled she had difficulty breathing. She often had to be admitted to hospital for observation. She has an affected sister, father, three uncles and one aunt. A vial of purified C₁ esterase inhibitor (C1-INH) concentrate was kept in the refrigerator for emergency use.

Stanozolol treatment was stopped as soon as she knew she was pregnant in April 1986. At 20 weeks gestation she developed erythema nodosa which settled over 2 weeks. A chest radiograph, white cell count and differential were all normal. She had four minor episodes of HAE which were no worse than before pregnancy. At 33 weeks she was admitted to hospital because of pre-eclampsia (blood pressure 160/100 mmHg, proteinuria and mild peripheral oedema). Thirty-six hours after admission she developed severe abdominal pain and vomiting, very unlike her HAE attacks, and required pethidine and anti-emetics. The uterus was initially soft and non-tender and there was no evidence of fetal compromise on the cardiotocograph (CTG). Seven hours after the onset of pain the uterus became tender and a repeat CTG showed episodes of fetal bradycardia. A diagnosis of placental abruption was made and an emergency cae-

sarean section was performed under epidural anaesthesia. A healthy 3.5 kg infant was born and at operation a 200 ml retroplacental clot was removed. The patient made an uneventful post-operative recovery and restarted stanozolol 6 weeks after delivery.

Patient 2

A 32-year-old woman had had recurrent episodes of abdominal pain associated with mild swelling of the limbs since adolescence. Her father was also affected. She had never had any prophylactic treatment. In 1982 she had an emergency caesarean section for failure to progress in the first stage of labour due to relative cephalopelvic disproportion. A healthy infant weighing 4.25 kg was born. During that pregnancy she had been admitted twice (at 21 and 33 weeks) because of abdominal pain needing analgesia and anti-emetics. In this pregnancy her HAE was much worse than usual, recurring every 5-10 days. At 13 weeks she was admitted for abdominal pain which settled with conservative management. At 23 weeks a very severe episode of abdominal pain unresponsive to analgesics was treated with 1000 units of purified C1-INH concentrate (Immuno Ltd, Arctic House, Rye Lane, Dunton Green, Sevenoaks, Kent) intravenously with relief of pain in 30 min. The remainder of the pregnancy was uneventful. A healthy infant weighing 3.58 kg was born after an elective lower-segment caesarean section at 39 weeks gestation under epidural analgesia. Two days postoperatively a further episode of severe abdominal pain unresponsive to analgesics was treated with 1500 units of C1-INH with good effect. Four days postoperatively danazol was started and at the 6 weeks postnatal check, the patient had had no further episodes of HAE.

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Discussion

Hereditary angioneurotic oedema (HAE) is a rare autosomal dominant disease (Landerman 1962) which can affect women during their reproductive years. It is characterized by recurrent episodes of subepithelial oedema resulting in swelling of tissues especially the face, larynx, limbs and gastrointestinal tract. The effect of pregnancy on HAE appears to be unpredictable, some reporting a benign course and others an exacerbation including one maternal death. The effect of HAE on the outcome of pregnancy itself has not been established.

HAE was originally described in the 1880s (Quincke 1882; Osler 1888). In 1963 a lack of C1-INH was found to be the cause (Donaldson & Evans 1963) and in 1965 two forms were differentiated, the predominant Type I (85%) due to a quantitative decrease in C1-INH and the less common Type II (15%) due to a functional decrease in C1-INH activity (Rosen *et al.* 1965).

HAE is inherited in an autosomal dominant fashion (Crowder & Crowder 1917). Recent identification of the C₁ inhibitor gene on chromosome II in the region p11.2-q13 has promoted further interest in the genetic basis for HAE (Davis *et al.* 1986; Bock *et al.* 1986). Use of recombinant-DNA probes for the C₁ inhibitor in the search of restriction fragment length polymorphism (Stoppa-Lyonnet *et al.* 1987) has demonstrated a structural gene defect. A DNA rearrangement resulting in both Type I and II forms is postulated, each family carrying an idiomorphic variant.

C1-INH is a glycosylated polypeptide with a molecular weight of approximately 108 kD and is normally present in high concentration in the plasma (17.4–24.0 mg/100 ml). It is the only known inhibitor of C1r and C1s, the activated proteases of the first component of complement (Gigli *et al.* 1968). It is also a major inhibitor of plasma kallikrein (Landerman *et al.* 1962) and activated Hageman factor (Ratnoff *et al.* 1969). Slow rates of inhibition of coagulation factors XIa and plasmin suggest that these are not significant *in vivo* (Travis & Salvesen 1983).

C1-INH acts by generating stable complexes of C1-INH with C1r and C1s, kallikrein, activated Hageman factor and plasmin (Laurell *et al.* 1978).

Serum levels of C1-INH fall in pregnancy because of an increase in plasma volume but the

total quantity is slightly increased. This is probably not functionally significant (Walker *et al.* 1982).

Deficiency of C1-INH results in chronic activation of the complement cascade with a resultant deficiency of C₄ and C₂ (Frank *et al.* 1976). The exact mechanism of activation of HAE attacks has not been fully assessed but anaphylatoxins C_{4a}, C_{3a}, C_{5a} and kinins are implicated.

Onset of attacks may be spontaneous or set off by emotional or physical trauma. First attacks usually occur in childhood or early adolescence: 50% will have their first attack before the 12th year, 5% will have their first attack after their 30th year (Frank *et al.* 1976). Frequency and severity of attacks diminish with age (Cicardi *et al.* 1982; Donaldson & Rosen 1966). A review of 72 affected patients showed that 93% of patients suffered recurrent abdominal pain, 96% peripheral oedema, 85% facial oedema and 64% pharyngeal oedema (Frank *et al.* 1976). In a review of 104 cases 56% of patients had recognizable prodromal symptoms and 18% had triggering factors (Cicardi *et al.* 1982). Episodes of HAE may be related to menstruation (Donaldson & Rosen 1966; Blohme *et al.* 1972; Frank *et al.* 1976). Attacks usually last 24–72 h but can range from 4 h to 1 week (Frank *et al.* 1976). Mortality, usually due to laryngeal obstruction, has been reported as high as 30% (Osler 1888; Donaldson & Rosen 1966).

HAE must be distinguished from other causes of pain and oedema. A careful family history and the recurrent nature of abdominal pain in association with peripheral oedema support the diagnosis but exclusion of surgical or obstetric causes may be difficult. Small bowel X-rays during attacks have shown mucosal oedema (Pearson *et al.* 1972) and unnecessary laparotomies have been performed (Cicardi *et al.* 1982). Toxic megacolon has been reported as a complication of HAE (Beck *et al.* 1973). Non-hereditary angioedema is more common and may be idiopathic or related to diet and may occur in association with septic shock, disseminated intravascular coagulation, malignant disease and drugs (e.g. Captopril). Anaphylaxis is differentiated from HAE by its rapid onset, non-recurrence and quick response to steroids, adrenaline and antihistamines.

Patients may be treated prophylactically especially if they have frequent laryngeal attacks. Antifibrinolytic agents have been used

to reduce the number of attacks, e.g. Epsilon-aminocaproic acid (Frank *et al.* 1972) by its direct action on C₁ activation (Lundh *et al.* 1968). Side-effects include muscle necrosis and a potential thrombotic effect; however, when tranexamic acid was used long term in 120 patients, no serious side-effects were noted (Agostoni *et al.* 1978). The potential thrombotic effect of antifibrinolytic agents makes it unwise to use in pregnancy though Logan & Greaves (1984) reported three pregnancies in one patient where acute attacks were treated with Epsilon-aminocaproic acid (4–6 g 6-hourly orally) without any side-effects.

Androgen derivatives, stanozolol or danazol, are better prophylactic agents (Gelfand *et al.* 1976) and work by increasing the synthesis of the inhibitor in the liver. There is a four- to fivefold rise in serum C1-INH activity. Long-term use of androgen derivatives is unwise unless the recurrent nature or severity justifies it. In pregnancy neither form of prophylaxis is recommended and both should therefore be used in conjunction with effective contraception and stopped when attempting to conceive. Masculinization of the fetus has been reported with patients on danazol (Hosea *et al.* 1980; Duck & Katayama 1981). One of our patients conceived whilst on stanozolol without apparent ill-effect.

Acute attacks are unresponsive to antihistamines, steroids or adrenaline and if they involve the larynx may result in death unless urgent supportive therapy is available. Purified C1-INH concentrate has been shown to be very effective in severe attacks (Bergamaschini *et al.* 1983; Agostoni *et al.* 1980; Gadeck *et al.* 1980; Logan & Greaves 1984). A subjective reduction of the airways obstruction and abdominal pain occurred in 30–60 min with the oedema subsiding over 2–3 h in 37 severe attacks described in these papers. Tracheotomy was never necessary. No side-effects or allergic responses have been noted. However, it is very expensive, there is a limited supply (on a named patient basis) and there is a potential risk of antibody formation, anaphylactic reactions and virus transmission. On the other hand it can be kept in the refrigerator at home and given by the general practitioner or a suitably trained person as soon as an attack starts. This is also specially useful for those who have infrequent attacks or where prophylaxis is contraindicated, e.g. pregnancy and childhood. Pre-operative infusion of C1-INH before endotracheal intubation may be a wise precaution.

Acute attacks have been treated with kallikrein inhibitor (Trasylol) but it is not as effective as purified C1-INH concentrate. Fresh frozen plasma contains C1-INH but should probably be avoided as it also theoretically provides the substrates which may be a source for the peptides of HAE. Lack of side-effects, efficacy and ease of use make C1-INH a superior form of treatment in the acute attack.

An increase in frequency of attacks in pregnancy has been reported in several case reports (Logan & Greaves 1984; Stiller *et al.* 1984) and only one where premature labour and possible stillbirth was attributed to HAE (Osler 1888). In a series of 25 pregnancies in affected patients only two had an increase in frequency in attacks and none of these was related to the delivery itself (Frank *et al.* 1976). Local oedema (infusion sites and vulva) has been noted after delivery (Hopkinson & Sutcliffe 1979; Stiller *et al.* 1984) but only one death due to shock 5 days post-partum has been reported (Postnikoff & Pritzker 1979). Vaginal delivery does not require any specific precautions.

Regional analgesia is preferable for operative deliveries to avoid the risk of laryngeal obstruction with endo-tracheal intubation (Hopkinson & Sutcliffe 1979).

Abnormal C1-INH function is detectable in affected offspring before the disease becomes manifest (Donaldson & Evans 1963). As effective prophylaxis and treatment of acute attacks is available, prenatal diagnosis using DNA gene probes appears to be inappropriate.

Conclusion

In pregnancy HAE runs a variable course and prophylaxis against attacks is unwise. Severe attacks should be treated with purified C1-INH concentrate. Episodes of abdominal pain and oedema must be carefully assessed and differentiated from obstetric and surgical causes. Vaginal delivery does not require any specific precaution but sites of local trauma (intravenous infusion, perineum, abdominal wound) should be carefully monitored. Regional analgesia is preferable for operative deliveries. Accessibility of C1-INH concentrate and communication between the supplier, patient, general practitioner, obstetrician, physician and anaesthetist are of paramount importance.

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Crohn's disease of the vulva. Case report

M. HOLOHAN, M. COUGHLAN, S. O'LOUGHLIN, P. DERVAN

Case report

A 35-year-old parous patient was referred to the gynaecology department with a 15-month history of painful, incapacitating ulceration of the anogenital and suprapubic areas and an offensive, locally irritating vaginal discharge. Investigations at the referring hospital had included bacteriological studies which had yielded only commensal organisms, vulval biopsy which had shown non-specific inflammatory changes and normal barium studies of the gastrointestinal tract. The patient had had partial relief from locally applied steroids without further improvement on additional oral steroids.

On examination there was a folliculitis of the vulval area with some shallow erosions and deep focal ulcers especially around the clitoris (Fig. 1). In the intergluteal cleft there were denuded inflamed areas.

Detailed bacteriological and fungal assessment did not yield any organisms considered to be of clinical importance. Haematological indices, biochemical screen, liver function tests, autoantibody screen, serology, glucose tolerance test and chest radiography were normal as was sigmoidoscopic examination. Serum complement fractions 3 and 4 were normal and serum IgE was not detected. The following, however, showed elevated values: C reactive protein 1.4 mg/dl (normal <0.6), properdin factor B 62 mg/dl (normal 17–42), IgM 572 mg/dl

(normal 50–250), IgA 1130 mg/dl (normal 140–500) and IgG 2060 mg/dl (normal 750–1650).

Biopsies from periclitoral ulcers showed surface ulceration with the residual squamous epithelium showing pseudoepitheliomatous hyperplasia. Granulation tissue was present in the superficial parts of the lesions and showed an intense—largely neutrophilic—inflammatory cell infiltrate. There was a background of histiocytes with occasional Langhan's giant cells and a few poorly formed granulomata. Ziehl-Neelsen and periodic acid Schiff stains were negative. No intracellular bodies were seen in the histiocytes or giant cells. Rectal mucosa was histologically normal.

Diagnoses considered were severe atypical intertrigo, Behçet's syndrome, tuberculosis and Crohn's disease.

Some improvement occurred with a 2-week course of oral tetracyclines and ketoconazole with local application of beta-methasone valerate, 0.1%, and dressings soaked in chlorinated lime and boric acid solution. The improvement was not maintained and over the following 3 months perianal skin tags appeared in conjunction with worsening ulceration. The skin tags were excised, and histological examination showed occasional non-caseating granulomata consistent with Crohn's disease (Gray *et al.* 1965). Fibreoptic and barium studies of the gastrointestinal tract were undertaken and were normal as was another rectal biopsy.

Treatment with metronidazole 20 mg/kg/day in three divided doses was started with striking improvement within 3 weeks. The folliculitis, erosions and ulcers healed completely but some vulval redness remained. The patient was asymptomatic. Therapy was discontinued after 6 months but within 2 weeks perianal ulceration had recurred and medication was recommenced. After therapy for a further year dosage was gradually reduced over 4 months. Off all treatment, the patient has now remained asymptomatic for 6 months.

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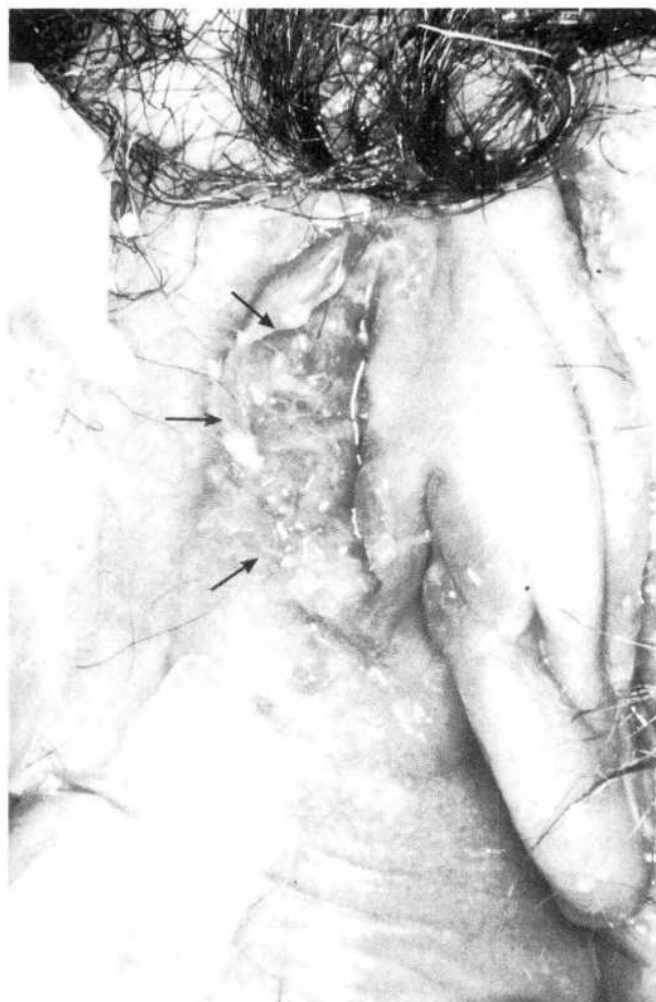


Fig. 1. Periclitoral ulceration in Crohn's disease of the vulva (edge of ulceration arrowed).

Discussion

Among the dermatological conditions associated with Crohn's disease, contiguous perianal lesions are common. Williams (1971) indicated that various authors have reported 11–93% incidence of perianal lesions in Crohn's disease. These lesions usually consist of indolent undermined ulcers showing granulomatous inflammation on microscopic examination associated with local abscess, fistula and fissure in the majority (Sutphen *et al.* 1984).

Freidrich (1983) stated that vulval ulcers may be the presenting feature of Crohn's disease in 25% of patients but he did not indicate whether this was ulceration alone or in association with abscess, fistula or fissure formation nor how

many had gastrointestinal disease when investigated. Spiro (1983) reported that perineal ulceration alone may rarely be the first sign of Crohn's disease and may precede any other manifestations of the disease by as long as 3 years. In a review of the gynaecological presentations of Crohn's disease, Donaldson (1978) found that 103 of 431 (24%) women with the disease had gynaecological features. Four of these presented with vulvovaginitis but all had fistula, fissure or abscess formation also and all except one developed gastrointestinal symptoms within 3 years of presentation. Bernstein *et al.* (1980) mentioned two patients who had never had gastrointestinal involvement but had perineal disease for 3 and 15 years respectively. Interestingly, it is reported that one of the

patients originally described by Crohn (1932) had labial ulcers for many years before developing bowel symptoms (Freidrich 1983).

The development of perineal skin tags as in our patient is an important clue to the diagnosis. It is noteworthy that in a patient reported by McCallum & Kinmont (1968), these skin tags resembled perineal condylomata.

Patients with Crohn's disease usually have normal immunoglobulins and complement components. Increased levels, as in our patient, are generally a marker for active bowel disease (Jewell 1983) which we did not find.

This patient's incomplete and transitory response to steroid medication but striking response to metronidazole is typical of the perineal manifestations of Crohn's disease. Marked improvement, if not complete healing, of such lesions occurred in all 21 patients treated with metronidazole by Bernstein *et al.* (1980) while therapy with steroids, sulphasalazine or broad spectrum antibiotics was not effective. The results following the use of immunosuppressant agents are contradictory and surgical intervention is followed by unhealed wounds in up to one half of patients (Corman *et al.* 1978).

Most patients tolerate metronidazole therapy without significant side-effects. A metallic taste and darkening of the urine are not uncommon but anorexia, vomiting and vertigo may also occur. There is a possible disulfiram-like effect with alcohol. Peripheral neuropathy may occasionally occur and may not completely reverse on discontinuation of therapy. Sudden discontinuation of therapy often results in recurrence of perineal disease, as in our patient, but tapering of the dose over 3–6 months before complete withdrawal may substantially limit the frequency of recurrence.

Conclusions

Vulval ulceration alone may be the presenting

feature of Crohn's disease. Perineal lesions may precede gastrointestinal disease by many years but more usually by less than 3 years. This emphasizes the need for continuing assessment of the gastrointestinal tract in patients who present with perineal disease alone. Metronidazole proves an effective therapy for the perineal lesions of Crohn's disease.

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Complete response to pre-operative chemotherapy in stage III B cervical carcinoma. Case report

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Case report

In January 1986 a 47-year-old white woman with five children was seen in the emergency room with severe haemorrhage from the upper vagina.

She was premenopausal and had regular periods but had had postcoital bleeding for 2 years. A cervical smear had not been performed in the past 7 years.

There was a large exophytic cancerous mass, $6 \times 5 \times 4$ cm, involving the cervix and the anterior and left lateral fornices. The body of the uterus felt normal. On rectal examination the left parametrium was involved to the pelvic wall but the contralateral parametrium was free of tumour.

Successful haemostasis was achieved by vaginal pack. The haemoglobin concentration was 10.1 g/dl. Punch biopsies consisted of squamous cell carcinoma of the small-cell poorly differentiated type. Examination under general anaesthesia and pelvic ultrasonography confirmed the volume of cervical mass and the involvement of the entire left parametrium while general examination, bone scan, chest X-ray, and hepatic ultrasonography were negative for distant metastases. Cystoscopy and proctoscopy were performed and no disease was detected. An intravenous urogram disclosed a marked hydroureteronephrosis on the left side due to obstruction of the ureter close to its entry into the bladder (Fig. 1).

Chemotherapy was given as primary treatment according to the following schedule: cisplatin 100 mg/m^2 i.v., day 1; bleomycin 15 i.u. i.v., days 1, 8; methotrexate 300 mg/m^2 i.v. plus cytrovorum factor rescue, day 8; repeated every 21 days, for three courses. The first course started on 23 January, and the last on 6 March, 1986 (day 1). Grade II-III nausea and vomiting and grade II alopecia occurred but there were no other side-effects.

Two weeks after the end of chemotherapy the patient was clinically restaged under anaesthesia in order to assess response. The cervix looked and felt normal and the induration in the left parametrium had resolved. Intravenous urography showed a dramatic improvement with only slight dilatation of the left ureter (Fig. 2). A cervical smear showed inflammatory changes, and colposcopy was negative.

On 28 March 1986, radical hysterectomy with radical para-aortic and pelvic lymphadenectomy was performed. Intra-operative blood loss of 1200 ml which was mainly due to injury of the right obturator vein during lymphadenectomy represented the only surgical complication, and did not substantially affect postoperative outcome.

On pathological examination, the cervix, corpus uteri, ovaries, parametria, vagina, paraaortic lymph nodes (caval: 6, intercavaortic: 14, aortic: 8) and pelvic lymph nodes (common iliac: 7, internal iliac: 12, external iliac: 15, obturator: 11) were reported to be free of malignant tissue.

The patient has remained in good health with no evidence of disease for 18 months.

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Discussion

Chemotherapy has been recognized as a possible treatment for patients with locally advanced cervical carcinoma. Published studies suggest a



Fig. 1. Pretreatment intravenous urogram showing left hydronephrosis due to stricture of the lower tract of the ureter close to its entry into the bladder.



Fig. 2. Intravenous urogram after the third course of chemotherapy; only a slight dilatation of the left ureter is visible.

clinical response in 30–70% of patients (Friedlander *et al.* 1984; Belinson *et al.* 1985; Kim *et al.* 1985; Vermorken 1986; Sardi *et al.* 1986) and a complete absence of tumour in some patients subsequently submitted to radical surgery (Kim *et al.* 1985; Sardi *et al.* 1986).

Our patient appears to have responded completely to chemotherapy which supports the findings of these workers. It is too soon, after only 18 months, to consider her cured but the rapid resolution of her advanced disease shows that pre-operative chemotherapy can be of considerable advantage in patients with advanced carcinoma of the cervix.

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Prolapse of the vaginal vault associated with a large granuloma of the sigmoid colon. Case report

R. W. A. NEL, J. T. NEL, N. CIROTA

Case report

A 69-year-old postmenopausal widow was admitted with a main complaint of a swelling protruding from her vagina. She also complained of a watery vaginal discharge. No stress incontinence of urine was present and her stools were completely normal. She had six adult children.

Fourteen years before the present admission she had a abdominal hysterectomy and bilateral salpingo-oophorectomy for postmenopausal bleeding, caused by endometrial polyps. Histological examination revealed no malignancy.

Four years before the present admission she had an operation for vaginal vault prolapse—the type of operation could not be ascertained. After this she had several cauterizations of granulation tissue forming in the vault of the vagina.

One year after the vaginal vault suspension she had a Burch colposuspension operation for stress incontinence of urine and anterior vaginal wall prolapse.

On general examination no abnormality could be detected. Her weight was 61 kg, blood pressure 160/90 mmHg, pulse 80/min and haemoglobin 12 g/dl.

There was complete prolapse of the vaginal vault covered by necrotic epithelium (Fig. 1). A

firm tumour could be palpated beneath the vaginal wall. Sigmoidoscopy was normal.

A cystogram showed good bladder filling with no reflux in the ureters. A moderate volume of residual urine was present after bladder emptying. No abdominal or pelvic masses were detected with ultrasound examination.

After pre-operative preparation the necrotic vaginal mucosa was dissected free of the underlying mass. The latter was found to be a part of the sigmoid colon containing a firm swelling (Fig. 2). Because of the likelihood of malignancy, the protruding mass was pushed back into the pelvis and at laparotomy the affected part of the sigmoid colon was resected. The proximal end of the resected colon was brought out on the anterior abdominal wall as an end colostomy and the distal end as a mucous fistula. The vagina was left open after removal of the necrotic tissue.

The resected colon was 16 cm in length and was partially encircled by a white tumour measuring $6 \times 5 \times 4$ cm. A further white tumour, $4 \times 3 \times 4$ cm, was present on the serosal surface of the bowel. Histologically there was prominent chronic inflammation with extensive immature fibrosis and granulation tissue formation. There were no signs of malignancy. The appearances were those of chronic inflammation of the prolapsed segment adjacent to the ulcerated vaginal wall.

The resected vagina measured $14 \times 8 \times 0.3$ cm. Microscopy revealed superficial ulceration with underlying acute on chronic inflammation, fibrosis and granulation tissue formation but no malignant changes.

The postoperative course was uncomplicated. The colostomy was closed after 4 weeks and, at the same time, the remaining vaginal mucosa and the pelvic floor was used to diminish the size of the vagina to try to prevent a further vaginal vault prolapse. Unfortunately, postoperatively the upper part of the vagina became necrotic.

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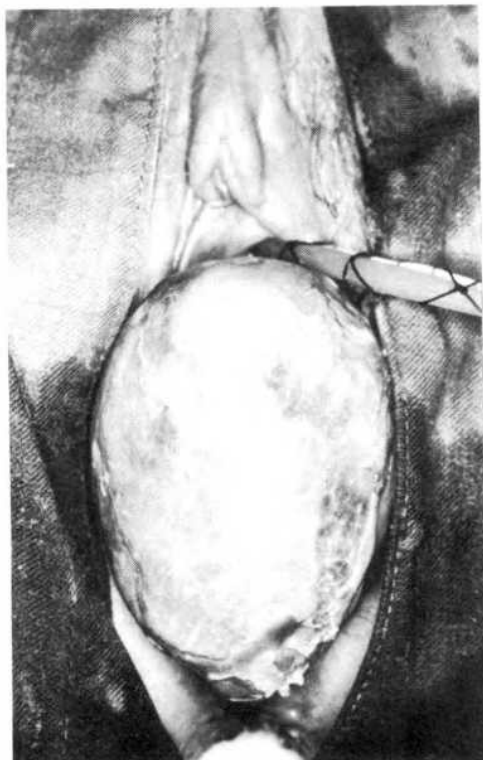


Fig. 1. Complete prolapse of the vaginal vault with necrotic vaginal epithelium.

Two months later this had healed but vaginal vault prolapse was again present. A colpocleisis was performed and at follow-up the result was satisfactory.

Discussion

Pseudotumours of the colon due to fibrosis following surgery of the abdomen or pelvis have been reported (Kyaw & Koehler 1972). The granulation tissue formation is, however, rarely so severe as to result in the formation of a tumour-like mass. The present case is, we believe, the first reported case of a large gran-



Fig. 2. Prolapsed sigmoid colon containing a firm swelling.

uloma of the sigmoid colon associated with vaginal vault prolapse.

Acknowledgment

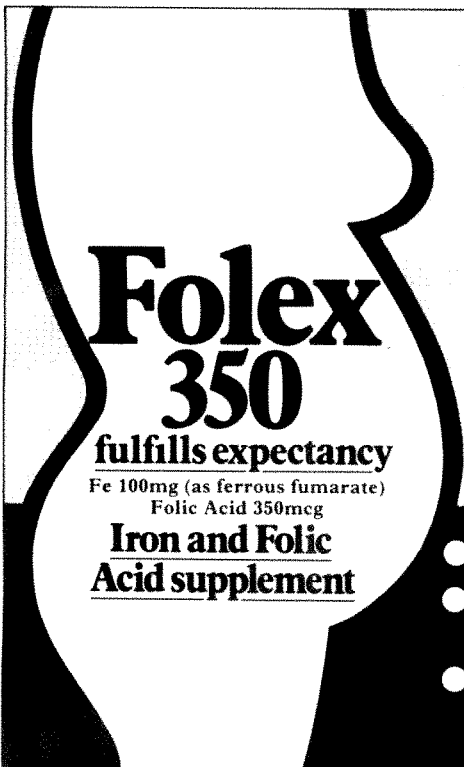
We wish to thank Mrs Cornia van Heerden for typing this manuscript.

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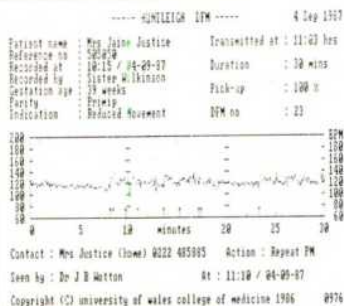
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Amoxycillin-clavulanic acid (Augmentin) versus metronidazole as prophylaxis in hysterectomy: a prospective randomized clinical trial

Dear Sir,

Dr Brown *et al.* [*Br J Obstet Gynaecol* (1988) **95**, 286-293] raised the question as to why antibiotic prophylaxis is effective in decreasing infective morbidity after hysterectomy irrespective of whether the chosen regimen has antibacterial activity against the aerobic or the anaerobic bacterial pathogens. Indeed they describe this as a paradox.

In a series of elegant experiments, Kelly (1980) demonstrated a pathogenic synergy between *Escherichia coli* and *Bacteroides fragilis* which helps to explain this phenomenon. When individually subinfective doses of these organisms were inoculated together into surgical wounds severe sepsis resulted. Similarly, when either anaerobes or aerobes were cultured from peri-operative wound swabs 13 to 22% of wounds became infected. When the peri-operative wound swabs cultured both anaerobes and aerobes the rate of postoperative wound infection was 71%. If either the anaerobic or the aerobic bacterial populations are decreased by antibiotic prophylaxis there will be a significant decrease in postoperative infection rates.

We have reported (Giles *et al.* 1984) our findings comparing antibiotic prophylaxis in abdominal hysterectomy, using metronidazole either alone or with cephadrine. Our findings and conclusions were similar to those of Brown *et al.* (1988) in that the use of metronidazole with cephadrine (with activity against both anaerobic and aerobic potential pathogens) effected a significant reduction in operative site infection compared with the use of metronidazole alone.

Thus, whereas inhibiting either the anaerobic or the aerobic bacterial flora provides an effective level of prophylaxis (probably preventing the synergy between the two groups of bacteria), an antibiotic regimen active against both groups

is still more effective in preventing postoperative infection.

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Concomitant zoonotic infections with ovine *Chlamydia* and 'Q' fever in pregnancy: clinical features, diagnosis, management and public health implications. Case report

Dear Sir,

The further case of *Chlamydia psittaci* infection in pregnancy reported by McGivern *et al.* [*Br J Obstet Gynaecol* (1988) **95**, 294-298] is, in fact, the ninth to date as two other confirmed cases have been reported from Edinburgh (Buxton 1986; Helm *et al.* 1987). More than half the reported cases have occurred in Scotland despite the fact that its sheep population is one-tenth that of the UK as a whole.

The authors make errors of detail, firstly in reporting that miscarriage occurred in five of the six cases reported before 1986, whereas miscarriage occurred in only one case at 25 weeks ges-

tation (McKinlay *et al.* 1985). Of the others, three went into premature labour at 23, 28 and 36 weeks gestation (Roberts *et al.* 1967; Beer *et al.* 1982; Johnson *et al.* 1985) and in the other, labour was induced at 28 weeks (McKinlay *et al.* 1985). In three of these five cases intrauterine fetal death occurred; in the case of spontaneous abortion the fetus died 2 h after delivery and in the other the time of fetal death is not reported.

Secondly, the authors state that in none of the previous cases was the organism specifically identified as an ovine strain of *C. psittaci* whereas both the isolate from the case reported by Buxton (1986) and that of Johnson *et al.* (1985) were specifically identified by restriction endonuclease analysis of their DNA (Herring *et al.* 1987).

The occurrence of *C. psittaci* early in pregnancy is worrying as this is now the second case to be reported (Buxton 1986) and raises the possibility of a role in early pregnancy loss which has previously gone unrecognized.

Since infection with *C. psittaci* is endemic within the sheep population, and its elimination is unlikely, the main effort should be directed towards preventing infection occurring in women who are pregnant, or who are trying to conceive. This is hampered by the fact that the exact route of infection is not yet known. The authors mention the possibility of transmission by fomites on working clothes and via airborne routes but they do not mention the possibility of infection via the genital tract or conjunctiva with contaminated hands and nails. With the present state of knowledge it would be best to advise women to avoid contact with *all* sheep while pregnant or trying to conceive, and not just those which are aborting or who have a history of aborting, and their partners should be advised to change contaminated clothes and wash all exposed parts before coming indoors after working with either healthy or diseased sheep.

Dr McGivern and his colleagues echo the need for wider publicity of the risks of sheep contact during pregnancy. These have already been circulated within the veterinary profession (Buxton 1986; Eddy & Martin 1986) and, indeed, one of the unfortunate women who suffered a severe infection was a veterinary surgeon. The risks are less well recognized by medical practitioners, particularly those in the community, to whom more information should be directed (Helm *et al.* 1987).

If more is not done then cases of infection

during pregnancy will continue to occur. The reported incidence of enzootic abortion of ewes within sheep flocks rose from 1110 in 1986 to 1448 in 1987 (Ministry of Agriculture, Fisheries and Food). To illustrate the potential scale of the problem there are 83 177 sheep farmers in the UK looking after 38 788 000 sheep, and counting children living on sheep farms there must be in excess of 100 000 women, just within the sheep-farming community, who are, were or will be at risk of infection.

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Dear Sir,

I would like to congratulate McGivern *et al.* [*Br J Obstet Gynaecol* (1988) **95**, 294-298] for once again drawing attention to the especial risk in pregnant women due to enzootic abortions of ewes caused by ovine *Chlamydia psittaci*. This is often neglected despite circulars from the Scottish Home and Health Department and news bulletins such as *The Times* newspaper (Prentice 1986).

I would, however, like to make several comments, namely:

- (1) That miscarriage in women may be caused by chlamydiae was first suggested by French workers in 1956 (Giroud *et al.* 1956).
- (2) The year of the reference 'Abortion due to infection with *Chlamydia psittaci* in a sheep farmer's wife' by Johnson *et al.* was misquoted throughout the whole paper, i.e. it should be 1985 instead of 1982. In later publications (Wong *et al.* 1985, 1986), the pathology of the chlamydial abortion due to severe placentitis has been well described. This was characterized by focal acute intervillitis which indicates the haematological route of spread of the infection. In addition, the chlamydial inclusions seen in trophoblasts were densely packed with elementary bodies, with less reticulate and intermediate forms. Various histochemical stains for glycogen did not demonstrate the presence of glycogen in these trophoblasts' inclusions, while inclusions due to *Chlamydia trachomatis* contained glycogen. This was further substantiated by transmission electron microscopy (Wong *et al.* 1985, 1986). In addition, Herring *et al.* (1987) had proved conclusively beyond any doubt that, by the restrictive endonuclease technique, the case by Johnson *et al.* (1985) was an abortion strain of ovine *C. psittaci* that had been acquired from the aborting ewe.
- (3) There was no mention of the differences among the different strains of ovine *C. psittaci*, namely arthritis, enteric or abortion strains. It is imperative that these strains are identified gen-

otypically by restriction enzymes technique for future epidemiological study as one of the main puzzles of the abortion strain of *C. psittaci* is its characteristic placentotrophism.

It was unfortunate that no fetal tissue, particularly the placenta, was submitted for histopathological and ultrastructural examination as this could help in understanding the pathology of chlamydial abortion.

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Authors' reply

Dear Sir,

We have read the letters by your correspondents Drs Helm and Smith and Dr Wong.

We apologize to the French workers (Giroud *et al.* 1956) for missing their paper recording their original observations on the role of *Chlamydia* in human abortions and for our error of nomenclature in referring to cases of premature labour and fetal loss as miscarriage.

Your correspondents raise points about the specific identification of *Chlamydia psittaci* of ovine origin in the case described by Johnson *et al.* (1985). We can merely reiterate that this *Chlamydia* isolate was *not* identified as an ovine *C. psittaci* strain in the paper published by Johnson *et al.* (1985). Subsequently Herring *et al.* (1987) identified this isolate as an ovine *C. psittaci* strain on the basis of restriction endonuclease analysis of chlamydial DNA. This paper was published *after* our paper was in press and this should have been obvious to your correspondents. We are, incidentally, unaware that the isolate reported by Buxton (1986) was identified as an ovine *C. psittaci* strain as this was not referenced in Dr Herring's paper. We suspect that part of this problem arises from repeat publications of isolates previously reported making it unclear as to the pedigree of different reports. May we take this opportunity of repeating that it was Dr Herring who kindly identified our isolate as an ovine strain by restriction endonuclease analysis.

Drs Helm and Smith comment on the mode of transmission which might have occurred in our case. We pointed out that a worrying aspect of this case was that the woman had no contact with sheep and we postulated that infection *may* have arisen as a result of her husband introducing the organism into the household from contamination of his clothing. This was speculative and we refrained from speculating even further to explain how she might have acquired the infection mechanically (as suggested by Drs Helm and Smith) in the absence of any acceptable scientific evidence. Other portals of entry could equally be postulated, i.e. the mouth and nose.

Finally, we emphasized that our patient acquired infection in the absence of clinical disease in the sheep and with no apparent contact and so it seems obvious that *all* sheep contact is a risk to a pregnant woman. We are pleased that Drs Helm and Smith concur with this view and for

the need for wider publicity to the medical profession.

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Reference ranges and sources of variation for indices of pulsed Doppler flow velocity waveforms from the uteroplacental and fetal circulation

Dear Sir,

With reference to your recent article [*Br J Obstet Gynaecol* (1988) **95**, 248-256], this provided some very useful information on the sources of variation when making Doppler flow velocity estimations, but with regard to the reference ranges, I would like to make several comments on the validity of your data.

Firstly, I was surprised at the inclusion of three patients (9%) who smoked cigarettes, and secondly, the use of data from a patient who developed fetal distress significant enough to warrant an abdominal delivery. Can the authors justify the inclusion of these four patients in a group of apparently 'normal patients'?

Lastly, was there any significant difference in the waveforms between the black, white and Asian patients? If so, such a heterogeneous group could not be used as a normal reference range for any individual patient, or indeed for an obstetric population such as our own, which is almost exclusively white.

P. P. Fogarty

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Reference

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Authors' reply

Dear Sir,

I thank Dr Fogarty for his comments on our paper as they give me an opportunity to bemoan the fact that clinical research, difficult at the best of times, is made more difficult by the intellectual disagreement, or in the case of reference ranges, sheer neglect from the epidemiological/statistical community. Most statisticians or statistical texts consulted before or since this study either gave no advice, produced unhelpful comments such as 'study as many patients who appear to be normal as you can', or gave examples illustrating the uselessness of such a range or the difficulty in defining a normal patient.

The philosophy that I therefore adopted in selecting and later excluding women used in this study was as follows. At the time the study was initiated it was becoming apparent that Doppler waveforms might be useful in patients with pre-eclampsia and/or intrauterine growth retardation. The lack of precision in the definitions of these end-points effectively meant that I had to be prepared to exclude any women who developed any degree or type of hypertension, and whose fetus or newborn infant was thought to be small by any definition! I selected patients that I hoped would prove to be a representative sample of the general antenatal population, hence the inclusion of smokers, and the failure to limit the study to any one racial group. I did not sample in a systematic way, for example every 100th woman, as I knew that I should receive many refusals because women have to work or care for other children. This is certainly not a random sample but it has ample precedent in that many other reference ranges are derived from hospital out-patients who happen to be available and free of apparent disease in the system under study; at least the 'patients' used in this study were healthy.

In view of the problems involved in creating a reference range, why not simply resort to significance testing or the use of confidence intervals to demonstrate a difference between the healthy and the diseased population? The answer is two-fold. Firstly, such an approach may be viewed as the result of an instantaneous window in time and is rarely helpful in the continuing clinical management of patients. Secondly, the frequency distribution curve from which such a range is derived should act as a starting point for

the study of why (and not just how often) individuals have particular values. From such a study we demonstrated that the resistance index from the uteroplacental circulation falls to a plateau at about 22–24 weeks gestation. This useful observation prompts speculation and further study into the underlying reasons.

More commonly, reference ranges are used as an aid to diagnosis in an individual. From the observation that values from normal patients rarely lie at the extremes we assume the converse, namely that it is common for values from diseased patients to lie at the extremes. This may be invalid and as such will make the test less sensitive. All investigators suffer from these limitations but if reference ranges demonstrate stability between populations (as is the case for the indices from the umbilical artery) then such data may more reasonably be referred to as a 'normal range'.

Having obtained a reference range, the next step is usually to determine whether the values vary systematically with individual characteristics such as height, weight, race, smoking habits and so on. In the vast majority of cases these characteristics are not relevant to the health of the individual but may well alter the values. I should argue that making allowances for the average effects of one (or all) of these characteristics is illogical because any effect that it may have on health is unknown and is likely to vary from individual to individual. Adjustment for these characteristics therefore merely introduces a further source of non-physiological variation such that even if all such characteristics could be recognized and allowed for, the remaining reference range would not represent physiological variation only. These comments obviously do not apply to the determination of experimental variation.

If Dr Fogarty is not satisfied with this philosophical answer, I can say that the data points from our cigarette smokers and the women who were delivered by caesarean section for fetal distress (whatever that means) are closely clustered about the midpoint of our frequency distribution. There were no statistically significant differences between the waveforms from the different racial groups but as the study was not designed to look for a difference, is this surprising?

Until our statistical/epidemiological colleagues squarely face the problem of reference

ranges and produce agreed guidelines I am afraid that Dr Fogarty will either have to accept my philosophy or create his own reference range.

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Prolonged maternal indomethacin therapy associated with oligohydramnios. Case reports

Dear Sir,

I agree with de Wit *et al.* [*Br J Obstet Gynaecol* (1988) **95**, 303-305] in their finding of oligohydramnios with prolonged indomethacin therapy. The authors did not feel that the oligohydramnios could be accounted for by decreased fetal urine output. I would, however, like to point out that indomethacin decreases urine output in the newborn (Cifuentes *et al.* 1979). Furthermore, Kirshon *et al.* (1988) demonstrated a significant decline in fetal urine output upon initiation of indomethacin for pre-term labour.

The mechanism of the decreased urine output from indomethacin is not only that of decreased renal perfusion but also via prostaglandin E_2 effect on antidiuretic hormone (Cifuentes *et al.* 1979). The preponderance of evidence suggests that class E prostaglandins antagonize the peripheral action of antidiuretic hormone (ADH) by blocking its stimulation of cyclic AMP (Anderson *et al.* 1976). Prostaglandins may also directly influence antidiuretic hormone release by a central effect (Gagnon *et al.* 1973). Inhibitors of prostaglandin synthesis may potentiate the effects of ADH (Anderson *et al.* 1975). Seyberth *et al.* (1983) demonstrated a rise in infant plasma ADH with initiation of indomethacin to affect ductal closure. This increase in ADH following prostaglandin synthesis inhibition is thought to

be the major factor resulting in decreased urine output.

While the authors state they noted the bladder to fill during indomethacin therapy, quantitative assessment of urine output was not performed. Based on Kirshon *et al.* (1988), I would suggest that the oligohydramnios from long-term indomethacin therapy resulted from decreased fetal urine output.

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BOOK REVIEWS

Essential Obstetrics and Gynaecology by E. Malcolm Symonds. Churchill Livingstone, Edinburgh, 1987, 266 pages illustrated, £14.95

This is an attractively illustrated and well-presented book which is a tribute to the energy and enthusiasm of its author who, since his appointment as Foundation Professor of Obstetrics and Gynaecology, University of Nottingham, has been a major force in the speciality in Britain.

He presents here what he considers to be the essential information required by medical students in their clinical course, which in some universities is a lamentably short period of 8 weeks. The book tries to focus on the common conditions encountered in clinical work and recognizes that while obstetrics has become more hospital-based, gynaecology is increasingly important in general practice.

The physiological basis of obstetrics and gynaecology is given in considerable detail. By contrast, some clinical topics are very briefly dealt with, although psychiatric disorders of pregnancy and the puerperium get a full chapter of 12 pages.

The book really consists of two sections, one on obstetrics and one on gynaecology, although there is some attempt at integration by dealing with the history and examination of the obstetric and the gynaecological patient consecutively in the first two chapters and by placing the consideration of infertility between 'placental and fetal growth and development' and 'complications of early pregnancy', where, to my mind, it sits rather uncomfortably.

The illustrations are delightfully clear and memorable, but perhaps the publisher overstates the case by claiming that the book is unique in this respect. *Obstetrics Illustrated*, for example, is a well-established publication in this style. One small criticism of the otherwise excellent illustrations is that obstetric patients are shown being examined naked in Chapter 1.

A minor irritation in the text is that 'Cusco's speculum' is consistently misspelt.

As the author of a different style of combined

book of almost identical name (now in its third edition) I welcome this new volume. A rose by any other name would, according to Shakespeare, smell as sweet, but whether two literary blooms should be marketed by the same publisher under indistinguishable names is open to question. But to change the metaphor—and the poet—"There are nine and sixty ways of constructing tribal lays, and every single one of them is right".

James Willocks

Premenstrual Syndrome by P. M. Shaughn O'Brien. Blackwell Scientific Publications, Oxford, 1987, 222 pages with figures and tables, £24.50

The understanding of the premenstrual syndrome has been bedevilled by so much bad work that Shaughn O'Brien is to be congratulated on producing a comprehensive assessment of available knowledge of this disorder.

It is in my view the only worthwhile book on the subject. It is comprehensive, well researched, well written, and as a sensible review of the muddle of PMS is certainly a spectacular achievement. The author has been very fair, even generous, in his assessment of conflicting views of aetiology and management, discussing them at times even when he is aware that the anecdotal uncontrolled nature of the claims (one hesitates to call it 'work') does not deserve to be taken too seriously or dignified by inclusion in this excellent book. Other writers would be less tolerant to those who have put the treatment of this common disabling condition on the wrong course for so long.

There are good chapters on the history and sociological and legal implications of PMS and also on the definition, the symptoms, the prevalence and epidemiology of the condition. The chapter on aetiology is a detailed examination of all of the traditional theories but, oddly, little mention is made of the role of ovulation in the causation of these cyclical symptoms or why

PMS becomes more severe as the menopause approaches nor is there any comment as to why the symptoms of PMS, with few exceptions e.g. mastalgia and violent behaviour, are also the symptoms of the menopause.

The chapter on therapy discusses the use of progesterone, progestogen, oral contraceptives, spironolactone, danazol, hormone implants and evening primrose oil. Herein lies the only weakness of the book in that in his desire to be objective, Dr O'Brien sits on the fence, unwilling to give clear advice on the correct treatment of the severe case who has no doubt already suffered many years of unhelpful therapy.

Treatment by suppression of the ovarian cycle with GnRH, danazol and oestradiol implants is well discussed. Although it seems clear to both the author and the reviewer that the only therapy which is more effective than placebo in every Moos cluster of symptoms is an oestradiol (not ethinyloestradiol) implant, he decorates his description of this technique with the strange caveat that oestradiol implants are much favoured by those who are in the private sector. God forbid that we should offer any of our patients anything but the best.

Dr O'Brien correctly states that PMS is not cured by hysterectomy with conservation of ovaries as cyclical symptoms persist. PMS is rarely, he states, an indication for 'bilateral oophorectomy'. That is true but he does not discuss the value of hysterectomy with bilateral oophorectomy in patients with intractable cyclical symptoms. This may be ideal primary therapy in selected patients with other menstrual pathology.

A similarly drastic therapy which should at least be an option given to women of appropriate age and family size is pregnancy. This is an important issue to address because effective therapy for severe PMS involves anovulation and a contraceptive state of some duration. Women may wish to complete their family before PMS compels them to throw in the towel.

These points are those which come to mind from my own view of the aetiology and treatment of premenstrual syndrome and in no way diminish my admiration for this book and my gratitude to Dr O'Brien for having spent a great deal of time writing it. It is a splendid book, full of good information, wisdom and controversy which deserves a large international market. We can only hope that Blackwells fulfil their role as a multinational publishing house and make a real

attempt at penetration of the North American market. My guess is that most anglophilic colleagues across the Atlantic will neither see a copy nor even have heard of it in years to come. It deserves better.

John Studd

The Health Scandal by Vernon Coleman. Sidgwick & Jackson, London, 1988, 245 pages, £12.95

The beginning of the nineteenth century witnessed the first age of therapeutic nihilism, when prophets of doom claimed that medicine had exhausted its usefulness and was instead posing a threat to survival. Coleman belongs to the second such age which began in the 1960s. Unfortunately, unlike some of his more thoughtful journeymen, Coleman's work is full of factual inaccuracies and contradictions and at times shows an ignorance and a lack of understanding of published literature. For example, we are told that between 1907 and 1977 'Life expectancy has gone up little, if at all'; 'half of NHS beds are occupied by stroke patients'; and that we could prevent most cancers, heart disease and strokes.

When he avoids such inaccuracies, he often ends up contradicting himself. Thus, whilst claiming 'medicine has virtually nothing to show for all the effort and expenditure' during this century, elsewhere the impact of cimetidine in halving the need for ulcer surgery is acknowledged; whilst reasonably citing the dangers of the restrictive practices of doctors, he suggests that they should replace health authorities and run the NHS with local politicians; whilst lambasting private medicine, he holds up the Sage renal dialysis case (treated privately) as a shining example of lay power; and whilst outraged by surgeons who refuse to operate on obese or smoking patients, he clamours for controls to be introduced on what we all should be allowed to eat.

For someone who claims and appears to have read widely, it is strange that he is only aware of one of the three major trials on coronary surgery and discusses the pertussis vaccine controversy without reference to the National Childhood Encephalopathy Study. And where the literature review is complete it is subjected to some doubtful interpretations: 2200 unnecessary deaths a year at the hands of surgeons assumes that unnecessary patients are at similar operative risk to necessary ones; and the lack of

impact that medicine has had on mortality is based on a false expectation—the principal roles of medicine are and always have been to reduce morbidity and disability.

Throughout, Coleman's style is to alarm his readers with his own unsubstantiated convictions: 'surgeons murder 10 000 people a year'; 'no one could argue that there is any less suffering in our society'; and 'stress is without doubt the major twentieth century killer'. The pity is that despite all the nonsense, Coleman manages to identify some of the problems that the medical profession poses to those who would try and improve the provision of care in Britain. What is lacking is any understanding of why change is so difficult. He seems to believe that it's all a conspiracy rather than a cock-up. In so doing he assigns the subjects of his criticism with more credit than they probably deserve.

Nicholas Black

Transvaginal Sonography edited by Ilan E. Timor-Tritsch & Shraga Rottem. Heinemann Medical Books Ltd, 1987, 165 pages with figures and tables, £29.95

This book describes the use of the vaginal ultrasound probe and speculates on its potential as a diagnostic aid.

The chapter which explains the principles of ultrasound imaging does not contain descriptions of other vaginal probes currently available, and the benefits or disadvantages of 90°, 105°, 135° angles have not been discussed. Thus, technical discussion is limited.

The methodology section explains vaginal ultrasound planes but some signs described such as 'the sliding organs sign' and tenderness on bimanual examination may be operator or patient dependent.

The three chapters on pelvic organ scanning deal with common conditions encountered in gynaecological practice; all have been described previously in gynaecological ultrasound books except that image quality is superior with vaginal ultrasound when compared to abdominal ultrasound. The authors have emphasized throughout that vaginal imaging is superior to abdominal ultrasound because of proximity of the transducer to the organs being imaged.

The chapters on early pregnancy scanning elaborate on the ability of vaginal ultrasound to detect structures and fetal heartbeat 1 week

earlier than with abdominal ultrasound. The importance of this is not discussed.

The diagnosis of ectopic pregnancy is shown to be possible with vaginal ultrasound, and the potential of this technique in diagnosing and treating ectopic pregnancies has brief mention.

In summary, this book has elaborated upon the improved images obtained by vaginal ultrasound and may be of use to ultrasonographers inexperienced in abdominal ultrasound. There is an abundance of ultrasound images.

For experienced radiologists and gynaecologists it does not contain information which is not available in other ultrasound textbooks.

Rajat K. Goswamy

Parasitic Infections in Pregnancy and the New-born edited by Caroline Macleod. Oxford University Press, 1988, 308 pages with figures and tables, £35.00

This is a useful reference book and, as far as I know, the first attempt to cover the topic in one volume. With the increase in worldwide travel, the subject is becoming of importance in Europe and North America in addition to the tropics, where most of these parasites are found.

Each chapter is set out in a similar way, beginning with basic facts about the parasite, its distribution and life-cycle. Next, the effects on the mother and fetus are considered, followed by the diagnosis and treatment during pregnancy. Each chapter concludes with an extensive list of references, no less than 14 pages in the case of malaria.

There are separate chapters on most of the important parasitic infections, including African and American trypanosomiasis, amoebiasis, giardiasis, toxoplasmosis, trichomoniasis and schistosomiasis. Malaria is comprehensively covered with a good discussion on the relative merits of different regimens of prophylaxis and treatment. Epidemiological and clinical aspects of trichomoniasis are discussed fully. Its prevalence is thought to be even greater in Third World countries than in Europe and the USA, but the high cost of metronidazole 'makes it unavailable to many women in developing countries' and there is as yet no suitable alternative. I was surprised to read that there was some evidence for a causal relation between gentian violet and malformation, so that even this old-established cure-all must be used with caution.

A chapter on 'The Pregnant Traveller' could

usefully be read by the traveller herself and contains some common-sense advice on avoiding illness in the tropics.

There are two invaluable appendices, the first of which summarizes all the obstetrician needs to know about the treatment of parasitic infections in pregnancy. The second lists the adverse effects of antiparasitic drugs on mother and fetus. Both teratogenic and mutagenic/carcinogenic effects are considered, and whether the drugs cross the placenta and are present in breast milk. The comments: 'no data', 'no studies', 'no evidence' and 'no reports' appear frequently, so it is not surprising that the editor in her preface appeals to readers to share their experiences and information for inclusion in future editions.

Jill Everett

Books received some of which may be reviewed at a later date

1988 Year Book of Obstetrics and Gynecology edited by Daniel R. Mishell Jr, Thomas H. Kirschbaum & C. Paul Morrow. Year Book Medical Publishers, Inc., Chicago, London, Boca Raton, 1988, 523 pages with figures and tables, £32.50

Voiding Function and Dysfunction: A Logical and Practical Approach by Alan J. Wein &

David M. Barratt. Year Book Medical Publishers, Inc., 1988, 371 pages, £28.50

Obstetrical Measurements in Ultrasound: A Reference Manual by Alfred B. Kirtz & Barry B. Goldberg. Year Book Medical Publishers, Inc., 1988, 214 pages with figures and tables, £34.00

Medical Complications During Pregnancy Third edition edited by Gerard N. Burow & Thomas F. Ferris. W.B. Saunders Company, Philadelphia, London, 1988, 603 pages with figures and tables, £43.95

Diabetes Mellitus in Pregnancy: Principles and Practice by E. A. Reece & D. R. Coustan. Churchill Livingstone, New York, Edinburgh, 1988, 651 pages with figures and tables, £65.00

The Menopause: Comprehensive Management Second edition edited by Bernard A. Eskin. Macmillan Publishing Company, New York, 1988, 350 pages with figures and tables, £32.50

Office Gynecology Third edition edited by Robert H. Glass. Williams & Wilkins, Baltimore, London, 1988, 362 pages with figures and tables, £48.00

Gestational Diabetes edited by Peter A. M. Weiss & Donald R. Coustan. Springer-Verlag, Vienna, New York, 1988, 241 pages with figures and tables, DM 138

Gynecologic Endocrinology and Infertility for the House Officer by Anne Colston Wentz, Carl M. Herbert III & George A. Hill. William & Wilkins, Baltimore, 1988, 303 pages, £10.50

Erratum

Br J Obstet Gynaecol 95, 621-623

Central pontine myelinolysis as a result of treatment of hyperemesis gravidarum. Case report
DAVID FRASER

The pretreatment level of urea quoted in the third paragraph was 86.5 mmol/l and not 8.65 mmol/l as stated in the Journal.